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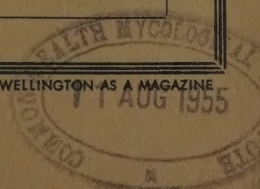
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CONTENTS

	PAGE
The Use of Chemical Thinning Sprays on Apple Trees in New Zealand. II. <i>R. M. Davison</i>	1
Growth of Pasture Species. II. <i>K. J. Mitchell</i>	8
Second Supplement to "A Revised List of Plant Diseases Recorded in New Zealand", 1952-1955. <i>R. M. Brien and Joan M. Dingley</i>	27
Methods for Determining the Viability of Various Seeds by Tetrazolium Staining. III. <i>E. O. C. Hyde</i>	36
Effect of Photoperiod on Varieties of Red Clover (<i>Trifolium pratense</i> L.) <i>L. W. Gorman</i>	40
Fertilizer, Cultivation, and Grassing Trials with Pampas Grass (<i>Cortaderia selloana</i>). <i>W. A. Jacques</i>	55
Screening Tests with Fungicides for Control of French-bean Rust (<i>Uromyces appendiculatus</i> (Pers.) Unger). <i>H. Jacks and R. M. Brien</i>	62
Adjustment of Casein/Fat Ratio of Milk for Cheesemaking. <i>R. M. Dolby and W. L. Harkness</i>	68
Publications Received	82

NOTICES

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June, 1955

Number 1

THE USE OF CHEMICAL THINNING SPRAYS ON
APPLE TREES IN NEW ZEALAND

PART II. FURTHER EXPERIMENTS WITH DINITRO
COMPOUNDS AND SYNTHETIC GROWTH SUBSTANCES

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(Received for publication, 9 March 1955)

Summary

Thinning sprays of sodium dinitro-ortho-cresylate (DNOC) and alpha-naphthalene acetic acid (ANA) on Sturmer in Hawke's Bay in 1952-53 were more effective than in previous seasons. Because of this factor and the lightness of the crop, overthinning occurred.

Moderately cropping Dougherty were not thinned as heavily as Sturmer. Both DNOC at full bloom and ANA at 50% petal fall showed promise as thinning treatments for this variety.

Tests on Sturmer in Nelson showed that several dinitro compounds and growth substances were effective thinning agents.

All ANA sprays and certain of the dinitro sprays increased russetting on both Sturmer and Dougherty. ANA on Sturmer in Hawke's Bay rendered foliage more susceptible to therapeutant-spray injury.

Chemical thinning sprays on both varieties in Hawke's Bay again induced a heavier blossoming in the spring following application. Sprays of ammonium dinitro-ortho-secondary-butyl phenate (DNBP) and sodium 2-methyl-4-chlorophenoxyacetate (MCPA) on Sturmer in Nelson, however, reduced the amount of blossom the following year.

INTRODUCTION

Results of experiments with chemical thinning sprays in the 1950-51 and 1951-52 seasons in Hawke's Bay and Nelson were recorded earlier (Davison, 1955).

During the season 1952-53 the two chemicals used in the previous trials, viz. alpha-naphthalene acetic acid (ANA) and sodium dinitro-ortho-cresylate (DNOC), together with other chemical thinning agents were tested in the same two districts. An account of these experiments is given below.

METHODS AND RESULTS

Experimental layout and method of recording were similar to those reported previously (Davison, 1955). In addition a bushel case of fruit taken at random from each tree was sorted to measure russet.

Chemicals used in these experiments were:

Dinitro Compounds

Sodium dinitro-ortho-cresylate (DNOC) as Selectone, 30% active chemical in water.

Ammonium dinitro-ortho-cresylate (ammonium DNOC), 30% active chemical in water.

Ammonium dinitro-ortho-secondary-butyl phenate (BNBP) as Selective-Selectone, 13% solution in water.

Dicyclohexylamine dinitro-ortho-cyclo-hexyl phenate as DN 111, a powder containing 20% active chemical.

Growth Substances

Alpha-naphthalene acetic acid (ANA) as contained in the pre-harvest-drop formulations Planofix and Phymone.

Alpha-naphthalene acetamide as the major constituent of the pre-harvest-drop formulation Fruitone. Fruitone also contains a minor proportion of ANA.

Sodium 2-methyl-4-chlorophenoxyacetate (MCPA) as Agroxone.

Hawke's Bay Experiments

On Sturmers, ANA used at two concentrations at petal fall was compared with a full-bloom spray of DNOC.

On Dougherty, ANA applied at 50% petal fall was compared with sprays of DNOC and DN 111 at full bloom.

Details of treatments and results are given in Table 1.

Thinning Effects

A moderate blossoming occurred on both Sturmer and Dougherty, and weather conditions over the blossom period were favourable for a satisfactory set of fruit. A heavy drop of young fruits from both varieties was observed in November before fruit-set counts were taken. As a result, Sturmers bore only a moderate crop necessitating only a light thinning. There was, however, a moderate to full crop on Dougherty.

On Sturmer all sprays overthinned, this effect being most pronounced with DNOC. The usual tendency to thin more fruit from the lower and inside parts of the trees was noted. Crop records show that all sprays increased fruit size, but fruit harvested from DNOC-treated trees was mainly oversize and undesirable.

On Dougherty, the lower concentration of both ANA and DNOC thinned satisfactorily over the upper parts of the trees but removed slightly too much fruit from the bases. The higher concentrations of these chemicals tended to overthin. DN 111 appeared to thin slightly but was not as effective as the other treatments. On this variety, only

the ANA sprays increased size. Fruit on Dougherty was generally small, and only the ANA 20 p.p.m. spray, which heavily overthinned, yielded fruit of a good size.

Subsequently all trees of both blocks were hand thinned where necessary.

Other Effects of Sprays

ANA and DNOC caused foliage injury on both varieties but no damage followed the DN 111 sprays. Leaves remained slightly inrolled

TABLE 1. EFFECT OF DINITRO AND GROWTH-SUBSTANCE THINNING SPRAYS ON STURMER AND DOUGHERTY APPLES IN HAWKE'S BAY, 1952-53

Treatment	Av. Yield/Tree Lb.	Log.	Av. Fruit Wt. (lb.)	Av. No. Fruit/ Case	No. Fruit/ 100 Clusters	XF Fruit* Per cent	Equiv. angle
<i>Sturmer</i> (5 trees per treatment)							
Unsprayed check	302.0	2.4674	0.262	154	67.3	34.4	35.62
Sodium DNOC 1½ pt./100 gal.	153.8	2.1741	0.420	95	21.4	45.9	42.65
15/10/52 Full bloom							
ANA 15 p.p.m.	263.3	2.4086	0.297	143	39.6	8.5	16.53
25/10/52 100% petal fall							
„ 20 „	250.6	2.3623	0.289	139	30.2	3.5	9.05
Diff. for sig.	5% 1%	0.2218	0.027	14.7	20.7		8.92
		0.3109	0.038				12.51
<i>Dougherty</i> (6 trees per treatment)							
Unsprayed check	353.3	2.5207	0.208	194	154.6	71.7	58.23
Sodium DNOC 1½ pt./100gal.	323.4	2.4340	0.221	186	94.3	61.7	51.86
13- 15/10/52							
Sodium DNOC 2 pt./100 gal.	203.2	2.2922	0.227	177	78.3	51.4	45.83
Full bloom							
DN 111 1 lb./100 gal.	264.9	2.3928	0.219	187	117.6	56.9	49.11
ANA 10 p.p.m.	268.4	2.4134	0.256	159	89.0	50.7	45.36
18/10/52 50% petal fall							
„ 20 „	152.1	2.1298	0.266	152	28.5	35.0	36.14
Diff. for sig.	5% 1%	0.2183	0.040	43.2	58.6		10.95
		0.2959	0.055				14.85

*The "Extra Fancy" (XF) grade included all fruit with less than 10% smooth russet and less than 3% rough russet.

Note.—(1) Full-bloom applications were made as follows: 1. Sturmer 80-90% blossoms open, 5-10% petal fall. 2. Dougherty 85-90% blossoms open, 5% petal fall, 5-10% late pink.

(2) Weather conditions over the blossom period were mostly partly cloudy to fine. Moderate winds were experienced over the two days preceding full-bloom applications. Unusually wet conditions prevailed in early spring and the season generally was wet.

and dwarfed following sprays of both concentrations of ANA on Sturmer and the 20 p.p.m. on Dougherty, the effects being most noticeable on less vigorous trees. Foliage injury caused by DNOC was only slight on Sturmer but was more apparent on Dougherty, being severe following the higher-concentration spray.

During the season, widespread marginal leaf scorch by sprays occurred on the Sturmer variety throughout the district. Injury of this type was noticeably more severe on trees treated with ANA sprays than on hand-thinned and DNOC-sprayed trees. This injury was more apparent on weaker trees. Foliage of Dougherty trees was undamaged by therapeutant sprays.

It was observed that on ANA-treated trees of both varieties a small number of partly developed fruits approximately one inch in diameter and containing abortive seeds remained attached throughout the season. These were not included in the crop but were included in the fruit-set counts.

Treatment with ANA caused a marked increase in russet on Sturmer as indicated in Table 1. A similar trend is shown on the Dougherty although russetting was not as severe as on Sturmer. DNOC at 2 pt./100 gal. also caused a slight but significant increase in the amount of russet on Dougherty.

No maturity differences in fruit were observed between any of the treated trees of either variety.

Trees of both Sturmer and Dougherty sprayed with ANA and DNOC developed a greater amount of blossom in the spring of 1953 than did hand-thinned check trees.

Nelson Experiment

Details of treatments applied to Sturmer and results obtained are given in Table 2. The commercial pre-harvest-drop formulations were used for thinning at the concentrations recommended by the makers for controlling fruit drop at harvest. Owing to unavoidable circumstances, DNBP was applied two days after the other full-bloom applications.

Thinning Effects

In the dinitro series all treatments except DN 111, which at the concentration used was ineffective, thinned significantly. Ammonium DNOC overthinned drastically.

All growth-substance formulations thinned effectively.

As unsprayed check trees set a moderate crop and required only a light thinning to break odd bunches, all effective sprays overthinned. Apart from a tendency to overthin the bases, distribution of the remaining fruit on trees following the majority of sprays was satisfactory. The spray of MCPA did not thin bunches effectively, but tended to eliminate fruit completely from some spurs thus giving an uneven distribution. This spray also kept small withered fruits attached until halfway through the season.

Other Effects of Sprays

Both dinitro compounds and growth substances caused foliage injury, the severity of which varied considerably with the compounds used.

TABLE 2. EFFECT OF DINITRO AND GROWTH-SUBSTANCE THINNING SPRAYS ON STURMER APPLES IN NELSON, 1952-53

Treatment (5 trees per treatment)	No. Fruit/ 100 Clusters	XF Fruit*	
		Per cent	Equiv. angle
Unsprayed check	113.9	35.5	36.26
Sodium DNOC 1½ pt./100 gal.	7/10/52 Full bloom	84.1	44.5
Ammonium DNOC 1½ pt./100 gal.		23.4	6.1
DN 111 1lb./100 gal.		103.1	24.9
DNBP 1½ pt./100 gal. 9/10/52		70.8	17.3
MCPA 20 p.p.m.		58.3	4.6
Phyomone to give 10 p.p.m. ANA	22/10/52 100% fall petal	69.9	10.9
Planofix to give 10 p.p.m. ANA		58.6	13.6
Fruitone 8 oz./100 gal.		84.8	31.4
Diff. for sig. 5%		22.4	10.54
1%		30.2	11.13
		Planofix	14.21
			15.07

*The "Extra Fancy" (XF) grade included all fruit with less than 10% smooth russet and less than 3% rough russet.

Note.—(1) Full-bloom applications were made when approximately 70% of clusters were fully open. Some petal fall had commenced.

(2) Weather over the blossom period was mostly cloudy and dry with cool nights.

In the dinitro series, severe injury resulted from application of ammonium DNOC and DNBP, trees damaged by these chemicals being slow to recover. Sodium DNOC caused only slight damage and DN 111 did not appear to cause any foliage injury.

Of the growth substances applied, MCPA alone affected foliage to any serious degree. This chemical caused severe curling and distortion of leaves and dying back of some shoot tips.

As in Hawke's Bay, therapeutant sprays applied throughout the season caused slight marginal leaf scorch, but in Nelson there was no evidence that ANA rendered foliage more susceptible to injury.

Ammonium DNOC and DNBP, of the dinitro series, both significantly increased the amount of russet. With the exception of Fruitone, growth-substance sprays also increased russetting.

No effect on maturity of fruit was observed.

The only treatments that affected flower-bud formation were MCPA and DNBP, both of which reduced the amount of blossom the following spring.

DISCUSSION

Results of these experiments give a further demonstration of the effectiveness of growth substances and dinitro compounds as thinning agents for apples in Hawke's Bay and Nelson.

On Hawke's Bay Sturmers, setting of 67.3 fruits per 100 clusters on the unsprayed trees for the 1952-53 season indicates that the crop carried was light, for in the previous two seasons the set, expressed on the same basis, was 142 and 90 fruits respectively.

The sprays had a proportionally greater effect than in 1951-52. For example, DNOC at $1\frac{1}{4}$ pt./100 gal. reduced set by approximately 68% compared with a reduction of 39% by a spray of $1\frac{1}{2}$ pt./100 gal. in 1951-52 (Davison, 1955). Batjer and Hoffman (1951) consider that thinning sprays are more effective under conditions that reduce tree vigour, e.g. water-logged soils in early spring. It seems likely that excessively wet soil may have influenced the results obtained with Sturmer in Hawke's Bay during 1952-53. The increased effectiveness of the thinning chemicals coupled with the lightness of the crop, both of which can be related to seasonal conditions, thus appear to be the main cause of overthinning on this block.

Apart from ANA 20 p.p.m. applied at 50% petal fall, the chemical treatments did not thin as severely on Dougherty as they did on Sturmer. Both DNOC and ANA, the latter applied at 50% petal fall, can be regarded as promising thinning agents for this variety. However, the foliage injury which followed application of the higher concentration of both chemicals is a factor to be considered.

Results of the Nelson experiment indicate that several dinitro compounds are effective thinning agents, but because of drastic overthinning and severe injury caused by certain of these sprays, more work is necessary to determine suitable concentrations. A point to be noted is the failure of DN 111 at 1 lb./100 gal. in both the Hawke's Bay and Nelson experiments. This chemical was reported as effective by Hoffman, Southwick, and Edgerton (1947), who considered also that it injured foliage less than sodium DNOC during humid seasons.

Results of this experiment and of previous experiments in 1951-52 suggest that MCPA cannot be regarded as a satisfactory thinning chemical, principally because of injurious effects and a tendency to reduce flower-bud formation. This conclusion is supported by observations of MCPA effects on a range of other varieties.

The smaller fruit size from the DNOC-sprayed Dougherty trees compared with fruit from ANA-sprayed trees in Hawke's Bay may have been brought about by foliage injury, offsetting the effects of early thinning. Dougherty appeared more susceptible to DNOC foliage injury than Sturmer.

As yields could not in all instances be correlated with the degree of thinning as shown by fruit-set counts, it seems likely that the smaller yields obtained from sprayed trees were largely due to the tendency of thinning agents to overthin the bases of trees. This emphasizes the need for concentrating thinning sprays mainly on the upper parts of trees during commercial applications.

An increase in russetting following sprays of dinitro compounds has been noted on a few occasions (Southwick and Weeks, 1949) but has not been considered serious. It is worthy of note that, in this respect, DNOC was safe on Sturmer and caused a significant increase in russetting on Dougherty only when the higher concentration of 2 pt./100 gal. was used.

A significant increase in russetting following growth-substance thinning sprays does not seem to have been reported previously, yet this effect was very marked with ANA on all blocks during 1952-53. Moreover, similar russetting on Sturmers thinned with ANA was noted on several other orchards in Hawke's Bay. Seasonal conditions undoubtedly were a major factor in determining the extent of russetting on the experimental blocks, since such effects of treatment were not observed in previous years. It is apparent, however, that under certain conditions thinning sprays, particularly ANA, could decrease the crop value of a russet-susceptible variety such as Sturmer.

It is evident from the observations made in Hawke's Bay that under some conditions ANA treatments tend to render foliage more susceptible to therapeutant-spray injury. The absence of additional injury of this type on treated trees in Nelson during 1952-53 and in previous experiments, suggests that this effect may be of consequence only under some climatic conditions.

Unlike the previous season, no discernible maturity differences between fruit on sprayed and unsprayed Sturmer trees were observed. However, subsequent storage experiments with fruit from this block will be reported in a later paper.

The effect of treatments on blossom formation of Sturmer and Dougherty in the following spring confirm the results of the 1951-52 experiments, and demonstrate that more regular blossoming and thus more regular cropping may result from the use of chemical thinning sprays on these two varieties in Hawke's Bay.

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GROWTH OF PASTURE SPECIES

II. PERENNIAL RYEGRASS (*Lolium perenne*), COCKSFOOT (*Dactylis glomerata*) AND PASPALUM (*Paspalum dilatatum*)

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(Received for publication, 1 February 1955)

Summary

The effects of temperature, defoliation, and shading on the pattern of vegetative development of young plants of perennial ryegrass, cocksfoot, and paspalum were determined. Plants were grown at mean temperatures of 59°F. and 83°F. At each temperature some plants were grown in full light and others shaded to 30% of full light. Some of the plants grown in full light were defoliated to approximately 6 cm. above ground level.

Under all conditions, except shading at 83°F., the increase in tiller number was faster with perennial ryegrass than with paspalum or cocksfoot.

For perennial ryegrass and cocksfoot, the rate of tissue formation by the primary tiller of undefoliated plants in full light was approximately the same at both temperatures, 5 to 6 mg. of dry matter per day. The rate was reduced by defoliation and by shading, the reduction due to defoliation being greater with cocksfoot than with ryegrass, and the reduction due to shading being relatively greater with ryegrass.

At 59°F. the rate of tissue formation by the primary tiller of paspalum was slower than for the other species, under all treatment conditions. At 83°F. it was much faster, reaching more than 25 mg. of dry matter per day with undefoliated plants.

The implications of differences in growth habit in relation to suitability of a species for different types of sward are considered.

INTRODUCTION

The first paper of this series reported a comparison of the patterns of growth of short-rotation and perennial ryegrass at different levels of light, temperature, and defoliation². The work was carried out in autumn 1952. The following spring similar comparisons were made among three species: perennial ryegrass, which grows vigorously in spring and autumn; cocksfoot, regarded as a summer producing species; and paspalum, which is confined to the warmer areas of the country and is a summer producer.

EXPERIMENTAL METHODS

The experimental procedure was similar to that of the previous experiment².

On 2 October 1952, seed of perennial ryegrass, cocksfoot, and paspalum was sown in a highly fertile potting soil in three-quart glazed earthenware pots. Ten days after emergence the plants in each pot were thinned to three per pot, selecting for uniformity within each species. The pots were then randomly allocated to the high and low temperature groups, means during the experiment being 83°F. and 59°F.

When there was a mean of 4 or 5 tillers on the plants in each species group, the pots at each temperature level were randomly allocated to

four sub-groups with 12 plants each. The first sub-group continued in full light, the second also continued in full light but was defoliated on that day and again seven days later, the third was shaded to 30% of full light but not defoliated, and the plants in the fourth were harvested for an estimate of weight of shoot at that time.

In order to ensure that the plants in each species group had commenced rapid initiation of tillers, the time of commencement of the treatments varied both in terms of stage of development of the plants as judged by the number of leaves formed on the main stem, and in terms of number of days from emergence. The mean number of leaves visible on the main stem was approximately 5.0 for perennial ryegrass, 6.0 for cocksfoot, and 6.7 for paspalum. The time taken to reach this stage of development varied from 20 days after emergence for perennial ryegrass at both levels of temperature, up to 36 days for paspalum at the low temperature.

The first defoliation removed all tissue above the ligule of the third leaf on the primary tiller, and the second all regrowth above the level of the previous cut. The height of cutting varied from 4 to 7 cm. above ground level, according to the habit of growth of individual plants.

The harvesting procedure, and the observations taken during the experiment, generally at two-day intervals, were the same as previously described.

Compared with the previous experiment² plants were grown to a greater stage of vegetative development, as judged by the number of leaves that had appeared on the main stem. For perennial ryegrass this averaged 9.5–10.0 for the various treatment groups, and for cocksfoot and paspalum approximately 11.0.

In considering the results, conclusions based on statistically tested data are drawn only where the test used indicates significance at least at the 5% level of probability. For other data only major differences are noted. Standard errors of the mean are normally given. Where results are obtained from interpolation or extrapolation from other data, the co-efficients of variation of the data from which the results are derived are given.

RESULTS

Growth of the Whole Plant

An estimate of weight of shoot tissue formed in the 15 days after final treatments commenced can be made, on the assumption that increase in shoot weight was logarithmic (Table 1). This integrates the effect on total shoot growth of the differences between species and treatments in tillering habit, leaf dimensions, rate of leaf appearance, etc. It can be seen that:

1. Defoliation and shading both reduced the total increase in dry weight of the shoot during this 15-day period by two-thirds or more.
2. Cocksfoot is estimated to have formed more tissue than perennial ryegrass where plants were undefoliated and in full light, but, if plants were defoliated, cocksfoot formed less tissue than perennial ryegrass. For cocksfoot, the smaller total increase in shoot weight at 83°F. than at 59°F. was due to initially smaller plants and not to a lower rate of growth.

TABLE 1. ESTIMATE OF INCREASE IN DRY WEIGHT (MG.) OF THE WHOLE SHOOT IN 15 DAYS
FROM THE START OF FINAL TREATMENTS

Treatment	Mean Weight of Shoot when Final Treatment Differences Commenced				Weight Remaining after First Defoliation				Estimated Weight of Tissue Formed in 15-day Period after Final Treatments Commenced*			
	Perennial Ryegrass 83° F. 59° F.	Cocksfoot 83° F. 59° F.	Paspalum 83° F. 59° F.		Perennial Ryegrass 83° F. 59° F.	Cocksfoot 83° F. 59° F.	Paspalum 83° F. 59° F.		Perennial Ryegrass 83° F. 59° F.	Cocksfoot 83° F. 59° F.	Paspalum 83° F. 59° F.	
Full light, undefoliated	46.0 45.0	28.5 42.7	43.5 43.0		— —	— —	— —		247 195	276 317	746 140	
Full light, defoliated	54.4 43.6	27.0 33.0	43.5 43.6		16.3 17.2	9.0 16.0	13.5 19.0		89 70	42 50	159 44	
Shaded, undefoliated	49.5 44.2	26.8 40.3	44.0 43.3		— —	— —	— —		83 69	55 81	198 48	
Co-efficients of variation	20% 22%	26% 19%	26% 22%		— —	— —	— —		25.35% 13.19%	25.35% 22.38%	15.24% 14.24%	

*Includes tissue removed in second defoliation.

3. At 83°F., paspalum in all treatments formed more tissue than the other two species, but at 59°F. it formed less tissue in all cases.

The ratio of root to shoot (Table 2) was highest on plants in full light at 59°F., averaging 0.44. It was reduced by increase in temperature or by shading. Differences between species were greatest with undefoliated plants in full light. Under these conditions, the proportion of root was lowest in cocksfoot and highest in perennial ryegrass.



Fig. 1.—Form of the shoot of perennial ryegrass. Full light, undefoliated at 83°F. (top left). Full light undefoliated at 59°F. (top right). Full light defoliated at 83°F. (middle left). Full light defoliated at 59°F. (middle right). Shaded to 30% of full light at 83°F. (bottom left). Shaded to 30% of full light at 59°F. (bottom right).



Fig. 2.—Form of the shoot of cocksfoot. Full light undefoliated at 83°F. (top left). Full light undefoliated at 59°F. (top right). Full light defoliated at 83°F. (middle left). Full light defoliated at 59°F. (middle right). Shaded to 30% of full light at 83°F. (bottom left). Shaded to 30% of full light at 59°F. (bottom right).

As leaf blades differ from sheaths in both structure and chemical composition, it is relevant to examine the proportions of each in the herbage (Fig. 4). Under all conditions *paspalum* formed relatively less blade tissue than either perennial ryegrass or cocksfoot. Usually, the proportion of blade tissue was increased by a rise in temperature or by shading, but not greatly affected by defoliation. At the extremes, the ratio of blade to sheath was approximately 1 : 1 with *paspalum* in full light, but 3 : 1 with the other two species when shaded at 83°F.

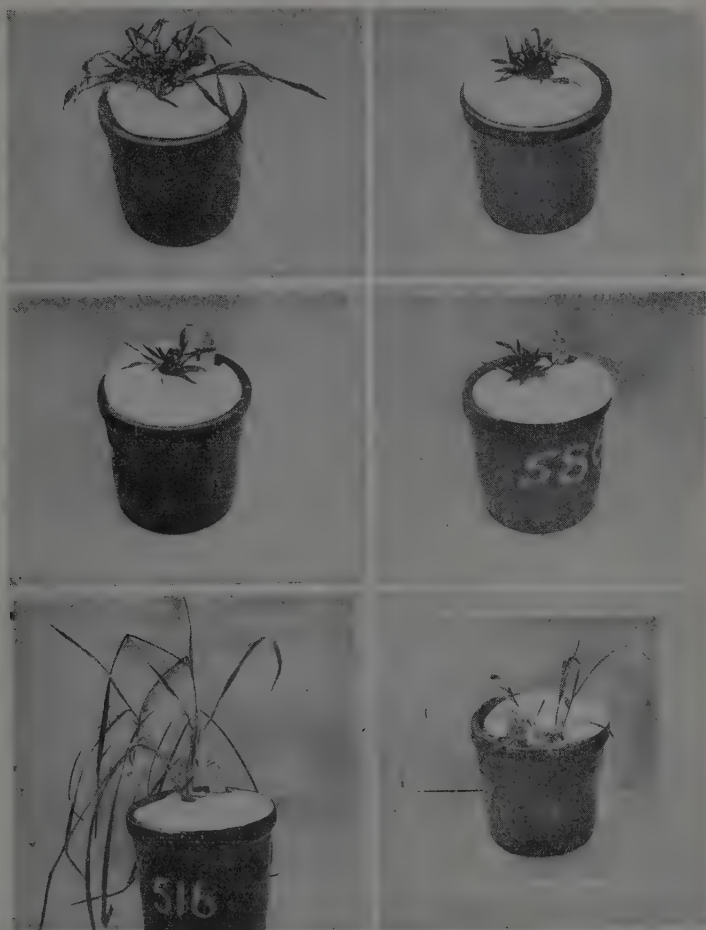


Fig. 3.—Form of the shoot of paspalum. Full light undefoliated at 83°F. (top left). Full light undefoliated at 59°F. (top right). Full light defoliated at 83°F. (middle left). Full light defoliated at 59°F. (middle right). Shaded to 30% of full light at 83°F. (bottom left). Shaded to 30% of full light at 59°F. (bottom right).

Tillering

Fig. 5 shows the effect of the treatments on the daily increase in the number of tillers. When the final treatments commenced there was a mean of approximately 4.0 tillers per plant for cocksfoot and paspalum and 5.0 for perennial ryegrass. The faster tillering of perennial ryegrass continued.

TABLE 2. RATIO OF ROOT TO SHOOT OF PLANTS AT THE TIME OF HARVEST

Species	Mean Temperature	Full Light Undeveloped S.E.	Full Light Defoliated S.E.	Shaded Undeveloped S.E.
Perennial ryegrass	83°F.	0.42 ± 0.02	0.25 ± 0.02	0.20 ± 0.01
	59°F.	0.54 ± 0.04	0.50 ± 0.05	0.15 ± 0.02
Cocksfoot	83°F.	0.28 ± 0.01	0.25 ± 0.01	0.17 ± 0.01
	59°F.	0.31 ± 0.01	0.34 ± 0.02	0.15 ± 0.01
Paspalum	83°F.	0.37 ± 0.04	0.27 ± 0.04	0.08 ± 0.01
	59°F.	0.49 ± 0.04	0.46 ± 0.01	0.32 ± 0.01

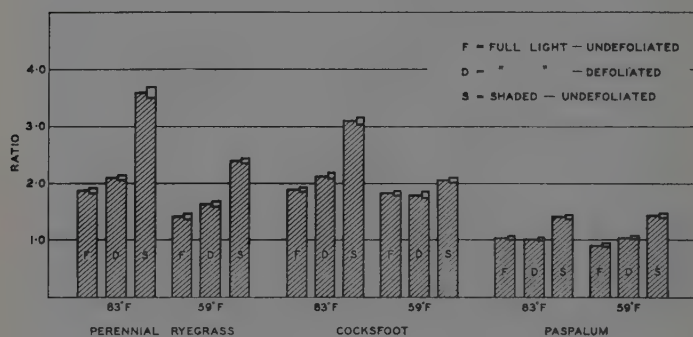


Fig. 4.—Ratio of leaf blades to leaf sheaths (dry weight).

In all species the number of tillers formed was decreased by defoliation and even more by continuous shading. For plants in full light, the rise in temperature from 59°F. to 83°F. had little effect on the number of tillers formed by perennial ryegrass but increased it a little for cocksfoot and very much for paspalum.

In both perennial ryegrass and paspalum, the reduction in tillering resulting from shading was greater at 83°F. than at 59°F., but, in cocksfoot, shaded plants continued to tiller at approximately the same rate at both levels of temperature.

The extent to which the potential of a plant for forming tillers is realized can be assessed by comparing tiller numbers when there are equal numbers of leaves on the main stem. This eliminates the effect of differences due to changes in rate of appearance of successive leaves on a tiller.

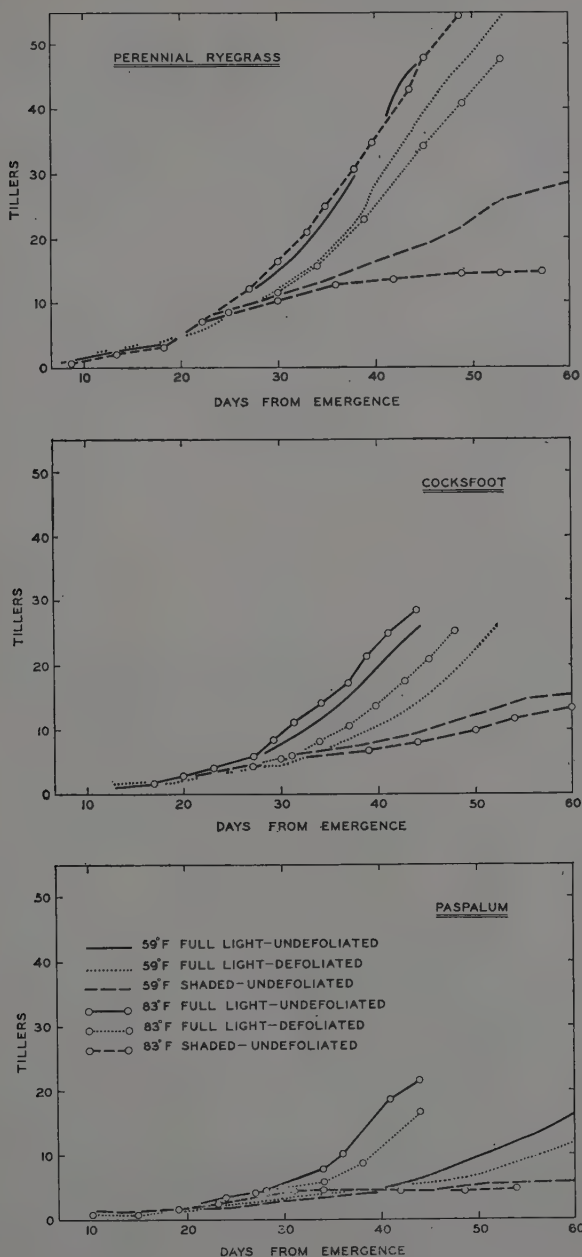


Fig. 5.—Daily increase in number of tillers per plant.

On this basis, marked differences between the species were apparent when there were 9.5 leaves on the main stem (Table 3). In full light, with or without defoliation, perennial ryegrass formed, in the period after final treatments commenced, 6 to 7 times as many tillers as paspalum, and 2 to 5 times as many as cocksfoot. Under shading, by far the greatest number of tillers (23.5) was formed by perennial ryegrass at 59°F., and fewest (0.6) by paspalum at 83°F.

TABLE 3. INCREASE IN NUMBER OF TILLERS PER PLANT IN THE PERIOD BETWEEN INITIATION OF FINAL TREATMENTS AND DEVELOPMENT OF 9.5 LEAVES ON MAIN STEM

Species	Mean Temperature	Full Light Undeveloped	Full Light Defoliated	Shaded Undeveloped	Co-efficients of Variation
Perennial ryegrass	83°F. 59°F.	44.0 50.7	33.7 44.9	9.0 23.5	19-30% 16-22%
Cocksfoot	83°F. 59°F.	11.5 10.3	13.1 10.6	6.4 6.1	17-30% 19-35%
Paspalum	83°F. 59°F.	7.0 6.9	5.1 6.0	0.6 2.2	16-22% 10-18%

The difference between perennial ryegrass and the other two species in number of leaves developed when treatment differences were initiated does not greatly affect the relative differences between these species.

The pattern of tiller formation in a ryegrass plant has been described in considerable detail¹ and observations during this experiment showed that it is similar for paspalum and cocksfoot. The primary tiller growing from the seed can be considered a parent. From its axillary buds first-order tillers arise; from these, second-order tillers; and so on. The relative contribution of tillers of the successive orders to the total weight of the shoot at harvest is shown in Fig. 6 for the plants grown at 83°F. Trends at 59°F. were similar but not so marked.

In perennial ryegrass in full light, both defoliated and undeveloped, about 60% of the total shoot weight came from third- and fourth-order tillers, i.e., from large numbers of young, hence comparatively small, tillers. The relative importance of the first- and second-order tillers is greater in cocksfoot and even more so in paspalum. In all species it is emphasized by shading. The extreme is reached in paspalum, shaded at 83°F., which had no tillers beyond first order and had 39% of its total shoot weight in the primary tiller.

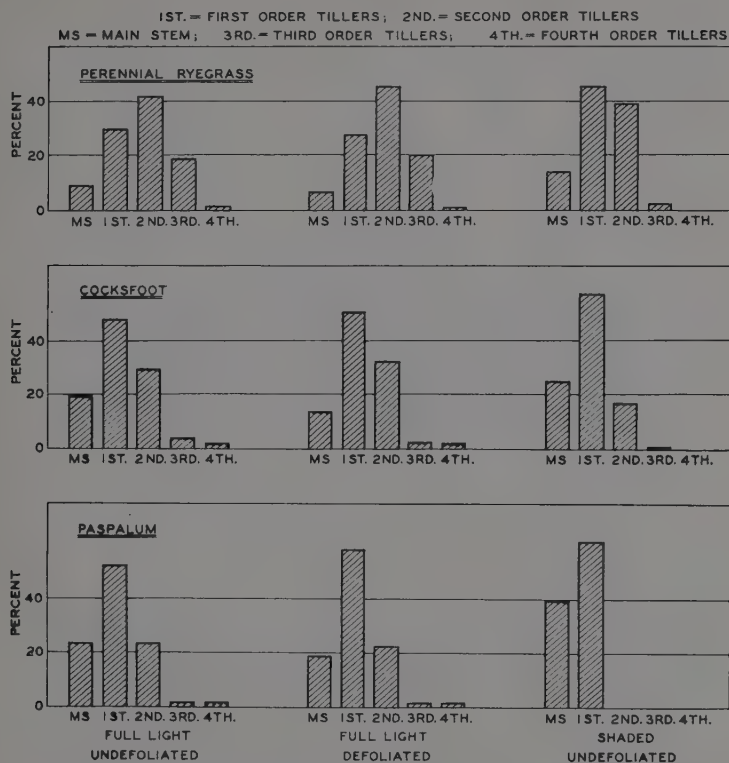


Fig. 6.—Percentage of total shoot weight present in the primary tiller and in first-, second-, third-, and fourth-order groups of tillers for plants grown at 83°F.

TABLE 4. AVERAGE WEIGHT PER TILLER
(mg. of dry matter)

Species	Mean Temperature	Full Light Undeveloped S.E.	Full Light Defoliated S.E.	Shaded Undeveloped S.E.
Perennial ryegrass	83°F.	30.4 ± 1.9	21.2 ± 2.5	48.3 ± 4.2
	59°F.	34.5 ± 2.0	14.3 ± 0.8	33.3 ± 1.3
Cocksfoot	83°F.	29.3 ± 2.8	7.2 ± 0.6	52.4 ± 4.1
	59°F.	25.7 ± 3.5	9.9 ± 0.9	36.6 ± 3.1
Paspalum	83°F.	58.6 ± 1.5	15.2 ± 1.5	208.4 ± 11.6
	59°F.	31.9 ± 2.2	15.3 ± 0.8	68.6 ± 3.5

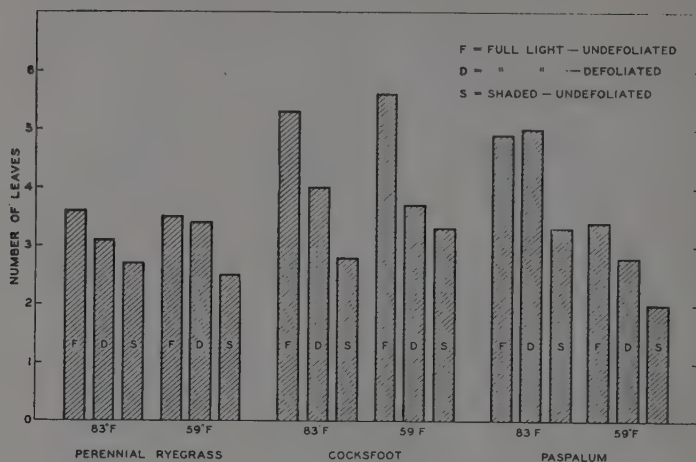


Fig. 7.—Mean number of leaves emerging on primary tiller in the twenty-day period after commencement of final treatment differences.

Average Weight per Tiller

Partly as a result of the absence of fourth-, third-, or even second-order tillers, average weight per tiller for the plant as a whole is usually higher in the shaded plants (Table 4). In some cases, e.g., paspalum and cocksfoot at 83°F., tiller weight of shaded plants is also increased by the formation of larger leaves (see Fig. 8).

Tiller weight was reduced by defoliation. The relative reduction was greatest with cocksfoot owing to a large reduction in leaf size.

Rate of Appearance of Successive Leaves on the Main Stem

The rate of appearance (Fig. 7) was similar at 59°F. and 83°F. in both ryegrass and cocksfoot but in paspalum was approximately a third lower at 59°F. than at 83°F. In all cases it was reduced by shading, but large reductions due to defoliation occurred only in cocksfoot. The range in mean number of leaves appearing in the 20-day period was from 5.6 in cocksfoot in full light at 59°F. down to 2.0 in paspalum shaded at 59°F.

Dimensions of Leaf Blade

Even within the same species, large differences in size and shape of the leaves may be induced by differences in temperature and light intensity or by defoliation. For example, in paspalum the blade of the ninth leaf on the main stem of defoliated plants at 59°F. had a mean weight of 4.1 mg. (dry weight) and a length of 3.2 cm. In plants shaded at 83°F. the comparable leaf blade weighed 59.2 mg. and was 43.4 cm. long.

Although data showing variations in dimensions are complex, they are relevant to so many issues that they merit recording.

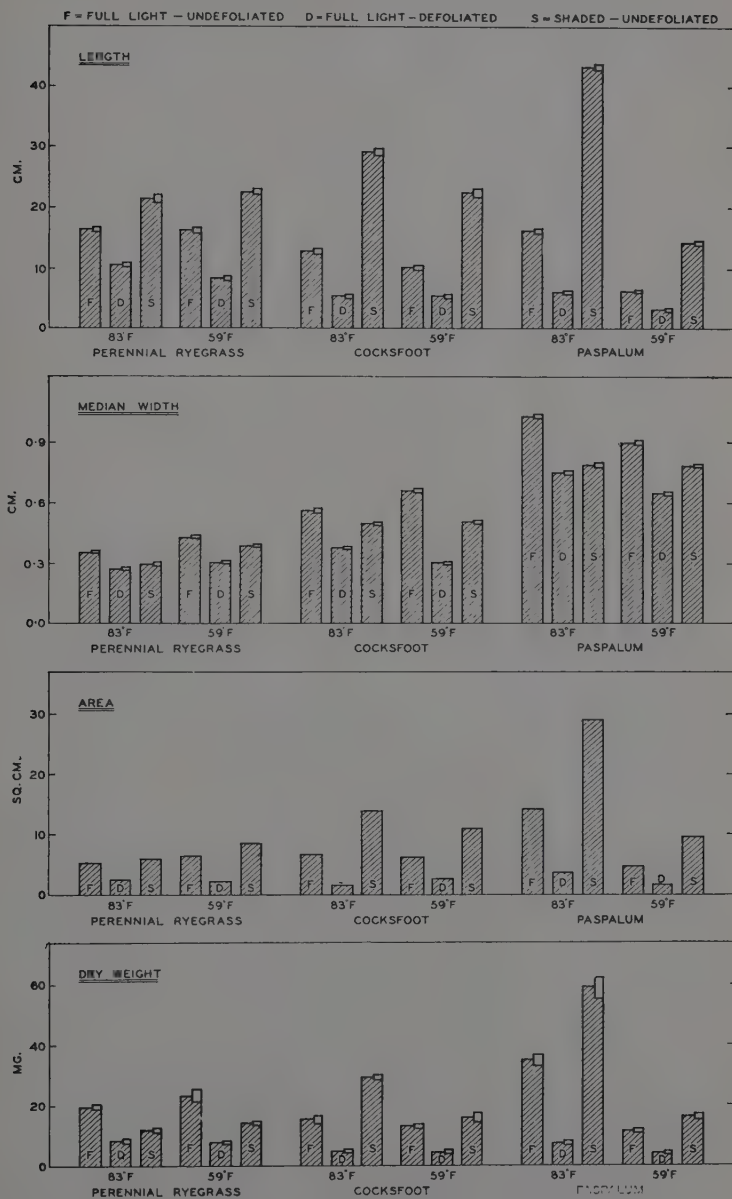


Fig. 8.—Dimensions of a representative leaf blade on the primary tiller.

In two defoliations six days apart, portions of several developing leaves on the main stem were cut off. The first intact leaf to appear subsequently on the main stem of the majority of plants was the seventh, eighth, and ninth for perennial ryegrass, cocksfoot, and paspalum, respectively. Comparisons of the effects of the treatments, set out in Figure 8 and Table 5, are based on these leaves.

TABLE 5. DRY WEIGHT PER SQ. CM. OF THE LEAF BLADE

Species	Mean Temperature	Full Light Undeveloped	Full Light Defoliated	Shaded Undeveloped
Perennial ryegrass	83°F.	3.45	3.10	1.99
	59°F.	3.90	3.17	1.68
Cocksfoot	83°F.	2.26	2.43	2.11
	59°F.	2.08	1.74	1.46
Paspalum	83°F.	2.46	1.98	2.04
	59°F.	2.41	2.33	1.73

In all species, defoliation reduced both the length and width, reducing the area of leaf blade in all cases by at least 50%. As it also generally reduced dry weight per sq. cm., the reduction in dry weight per leaf blade from defoliation was even greater.

Leaf blades of shaded undeveloped plants of perennial ryegrass increased in length by approximately a third and those of paspalum and cocksfoot more than doubled in length. In some cases this was partly offset by a reduction in blade width. The resultant increases in leaf-blade area are not fully reflected in weight, for shading in all cases reduced the weight per square centimetre of the blades. With perennial ryegrass the total weight of the leaf blades was less than on undeveloped plants grown in full light.

Whether the leaves of one species were longer or shorter than the leaves of another depended on the treatment, but in all cases leaf-blade width was least in perennial ryegrass and greatest in paspalum. Weight of leaf blade per square centimetre was about 25% greater in perennial ryegrass than in paspalum or cocksfoot with plants in full light, but in shade there was less difference between the species. The rise in mean temperature gave a large increase in the weight of blades of paspalum, a smaller increase in cocksfoot, and little effect in perennial ryegrass.

Rate of Tissue Formation by the Primary Tiller

This was calculated in the same manner as in the previous experiments¹. Short term fluctuations were minimized by two cycles of unweighted five-point smoothing.

The influence of species and treatment on the amount of tissue formed on successive days during the trial is shown in Fig. 9, and Table 6 sets out the total quantity of tissue formed in the 15-day period after commencement of final treatments.

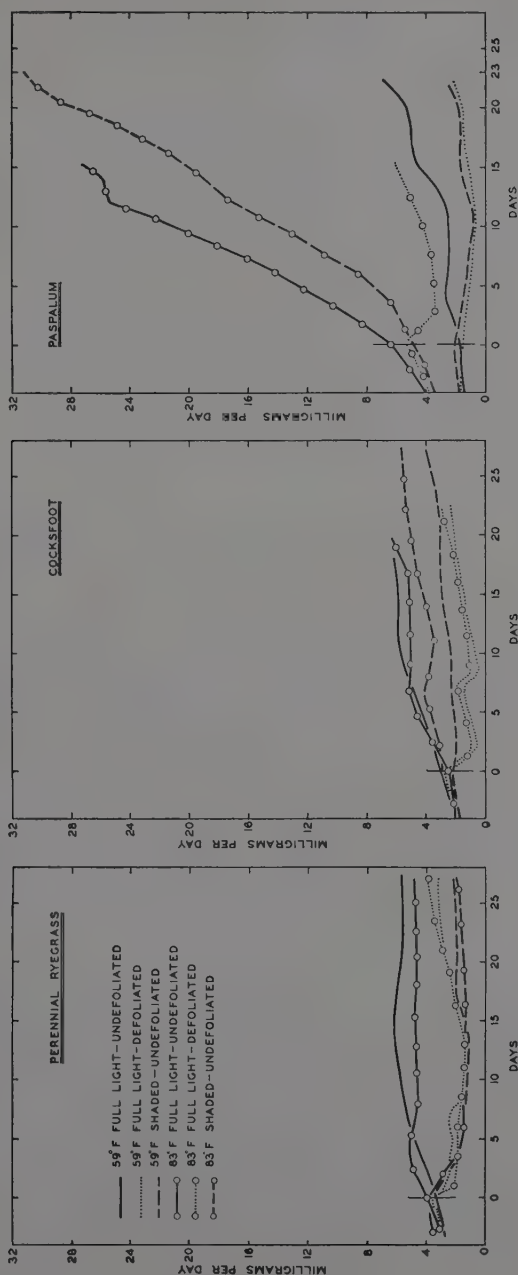


Fig. 9.—Estimated quantity of tissue formed each day on the primary tiller. The date on which the final treatment differences were initiated is taken as day zero.

TABLE 6. TOTAL QUANTITY OF TISSUE FORMED ON THE MAIN STEM TILLER IN 15 DAYS AFTER TREATMENTS (mg. of dry matter)

Species	Mean Temperature	Full Light Undeveloped	Full Light Defoliated	Shaded Undeveloped
Perennial ryegrass	83°F. 59°F.	71.0 77.4	27.5 29.6	27.7 24.6
Cocksfoot	83°F. 59°F.	72.0 75.6	21.0 15.1	52.5 35.2
Paspalum	83°F. 59°F.	265.6 44.1	62.7 16.5	181.4 20.8

Relative quantity of tissue formed. Full light—Undeveloped taken as base 100

Perennial ryegrass	83°F. 59°F.	100 100	39 38	39 32
Cocksfoot	83°F. 59°F.	100 100	29 20	73 46
Paspalum	83°F. 59°F.	100 100	24 37	68 47

For plants grown in full light without defoliation the following points can be noted:

1. In perennial ryegrass, after an initial period in which there was a continued increase in the quantity of tissue formed on successive days, the primary tiller formed tissue at an approximately constant rate per day. This was about 5.0 mg. D. M. per day for plants at 83°F. and 6.0 mg. at 59°F. An initially higher rate of tissue formation at 83°F. was offset by an earlier levelling off. Hence, the weight of tissue formed in the 15 days after final treatment differences commenced was approximately the same at both levels of temperature.
2. In cocksfoot the daily rate of formation of tissue by the primary tiller was initially lower than in ryegrass, but the trend was for it to continue to increase throughout the trial. The difference in temperature had little influence on quantity of tissue formed by the tiller. Also, over the 15-day period it was approximately the same as in perennial ryegrass.
3. In paspalum the quantity of tissue formed each day by the primary tiller was much greater at 83°F. than at 59°F. At both levels of temperature it continued to increase throughout the experiment and in plants at 83°F. had risen to 26 mg. per day at the time the plants were harvested. Over the 15-day period paspalum grown at 83°F. formed over three times as much tissue on the primary tiller as did perennial ryegrass or cocksfoot, but at 59°F. paspalum formed little more than half as much tissue.

Where plants were defoliated, the quantity of tissue formed by the primary tiller in the fifteen-day period, was, in all cases, reduced to less than 40% of that formed by undefoliated plants in full light. It will be noted that:

1. The relative reduction in weight of tissue formed was least with perennial ryegrass.
2. Even 12 to 15 days after the second defoliation the primary tillers of defoliated plants were still forming much less tissue per day than were equivalent tillers on undefoliated plants in full light.
3. In perennial ryegrass and cocksfoot, but not paspalum, the daily rate of tissue formation by the tiller of defoliated plants was similar at both levels of temperature.

The relative reduction in quantity of tissue formed by the primary tiller resulting from the shading to 30% of full light was approximately the same at both levels of temperature in perennial ryegrass, a reduction of nearly two-thirds compared with undefoliated plants in full light. However, in both cocksfoot and paspalum the shading gave relatively less reduction. This was particularly so at 83°F. where the quantity of tissue formed fell by less than a third in both species.

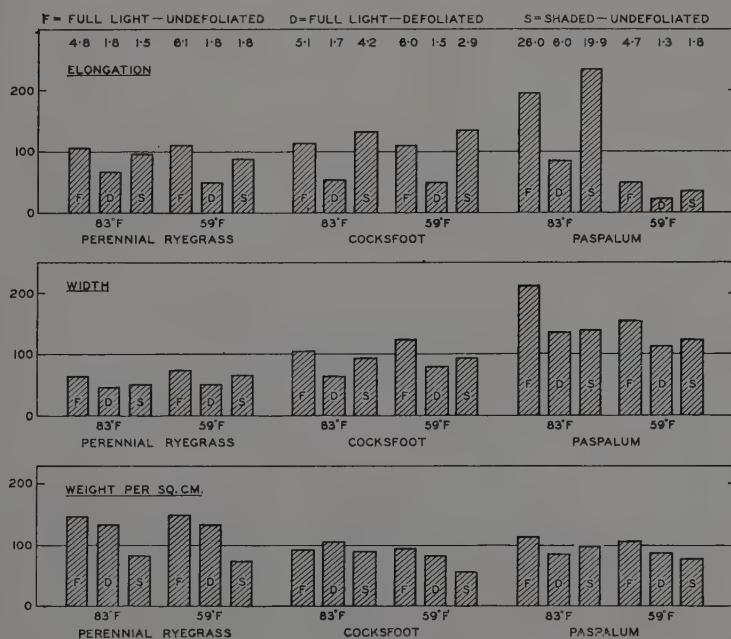


Fig. 10.—Relative importance of rate of leaf elongation, leaf width, and weight per sq. cm., in determining quantity of tissue formed on the fifteenth day after commencement of final treatment differences. The mean for all groups is base 100. The quantity of tissue estimated to have been formed on the fifteenth day is shown at the top.

The average of each of the above factors for all treatments and all species is taken as base 100 to give an indication of variation in each as between species and treatments.

As stated above, the daily rate of tissue formation on a tiller was calculated by multiplying together daily increase in length of the growing leaves on the tiller, the mean width of the leaves being formed, and their dry weight per square centimetre. The relative importance of each of these factors in determining the weight of tissue formed by the primary tiller on the 15th day after the final treatments commenced is shown in Fig. 10.

The very rapid rate of tissue formation by a tiller of *paspalum* at 83°F. was due to fast elongation of wide leaves. In perennial ryegrass, the rate of elongation of leaves on the tiller was in all treatments of the same order as in cocksfoot. The leaves being formed by perennial ryegrass were narrower but of higher dry weight per square centimetre. In all species a concurrent fall in leaf width and rate of elongation was mainly responsible for the slower growth by the individual tiller when defoliated.

DISCUSSION

Major features of the growth of each species may be summarized as follows. Under all conditions, except shading at 83°F., the increase in tiller number was faster in perennial ryegrass than in cocksfoot or *paspalum*.

In undefoliated plants in full light, the rate of tissue formation on the primary tiller was approximately the same for perennial ryegrass and cocksfoot. Compared with perennial ryegrass, the reduction in quantity of tissue formed by the individual tiller of cocksfoot was greater when the plants were defoliated and less when shaded. At 59°F. the rate of tissue formation by a tiller of *paspalum* was slower than in other species. At 83°F. it was much faster.

The implications of these differences in tillering, in quantity of tissue formed by the individual tiller, and in the associated changes in leaf dimensions, may be considered in relation to three sets of pasture conditions. Broadly, the double-defoliation treatment would resemble conditions in a closely defoliated sward: the full light without defoliation, a sward held at a moderate length, viz. 4 to 6 in. long; and the shading treatment a sward allowed to grow long.

High production from a closely defoliated sward would depend on there being large numbers of tillers per unit area to offset the relatively small weight of tissue formed by each tiller. Rapid formation of new tillers, despite the close defoliation, would allow prompt replacement of casualties from trampling, etc., and the formation of narrow thick leaves should give advantage by minimizing mutual shading.

At low temperature, perennial ryegrass met these requirements best. Under defoliation it tillered fastest, formed most tissue on the individual tiller, and had highest dry weight per square centimetre of leaf blade. At the higher temperature, the reduction in tillering of defoliated plants of perennial ryegrass, and also the considerable fall in proportion of root, suggest that its vigour of growth was commencing

to be adversely affected. On all counts cocksfoot still appeared inferior to perennial ryegrass, while the individual tiller of *paspalum* formed tissue more than twice as fast as perennial ryegrass by producing a rapid succession of small leaves. This does not automatically imply an equal rise in the production per unit area of ground from a closely defoliated sward. *Paspalum* formed wider leaves, 0.75 cm., against 0.27 cm. for perennial ryegrass, and consequently each tiller would take up a larger area of ground, partly because of its increased basal diameter, and partly because of the considerable mutual shading from such wide leaves.

In a medium-length sward a high rate of tillering could become of less importance, for tiller casualties should be less and there is no longer the need to fit in a maximum number of small vigorous tillers to ensure complete ground cover. The rate of tissue production from each tiller present should be relatively little checked by either defoliation or shading.

In terms of both production from the individual tiller and rate of tillering, there appeared to be little to choose between perennial ryegrass and cocksfoot, at either level of temperature, for plants grown undefoliated in full light. At the higher temperature the individual tiller of *paspalum* formed tissue nearly four times as fast as either perennial ryegrass or cocksfoot. The plant was at the same time able to tiller well and form a good proportion of root. Once again, large leaf dimensions should prevent this from being fully reflected in herbage production from an area of sward, by reducing the number of tillers that can be fitted in. At the lower temperature, both rate of tillering and growth from the individual tiller were less in *paspalum* than in cocksfoot or ryegrass.

Although these are the logical conclusions from the data, they illustrate the caution needed in their direct application to field conditions. For instance, the difficulty of maintaining cocksfoot in the freely tillering condition, in a medium length pasture, without having the plants either developing to a coarse tuft or being severely checked by close defoliation is well known.

Where a sward is grown to considerable length, the high degree of mutual shading between plants severely restricts tillering by plants of most species. The emphasis changes to ability to form a large amount of tissue on those tillers present, and ability to grow leaves of sufficient length to avoid overshadowing by adjacent plants. Such requirements are generally met by large tillers.

At the lower temperature level, there appeared little difference between perennial ryegrass and cocksfoot when shaded. Leaf length was about equal, and the higher rate of tissue formation by a tiller of cocksfoot could be balanced firstly by the faster tillering of ryegrass and secondly by the smaller, thicker, leaves of perennial ryegrass, which should enable it to maintain more tillers per unit area of ground. In all respects the growth of *paspalum* was poorer.

At the higher level of temperature the relationships between the species under shading were very different. With the rise in temperature from a mean of 59°F. to 83°F., the weight of tissue formed by a tiller of perennial ryegrass was unchanged, but with *paspalum* it was increased ninefold. At 83°F. mean temperature, the rate of tissue formation by the tiller of *paspalum* was three times as great as that of cocksfoot, which

in its turn formed tissue twice as fast as perennial ryegrass. The species rank in the same order in terms of leaf length.

The ability to develop large tillers, which give high production despite considerable mutual shading, will often necessitate increased care in utilizing that growth. The low rate of tiller reproduction gives a limited population of young tillers rooted and able to replace casualties either during growth or at the time of grazing or mowing. Uniformity of leaf length becomes much more important, otherwise shading competition will check plants with the shorter leaves, eventually leaving gaps in a pasture, which are often not readily filled by desirable species. Also, from its very size, and from the low proportion of root formed, when shaded, the large tiller may be more susceptible to trampling damage, pulling out by stock grazing, or killing due to sudden drying out and heating of the soil surface when exposed by grazing or mowing.

ACKNOWLEDGEMENTS

The assistance of Mr. R. A. Waters and Mrs. P. D. Fejer in carrying out these experiments is gratefully acknowledged.

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SECOND SUPPLEMENT TO "A REVISED LIST OF PLANT DISEASES RECORDED IN NEW ZEALAND", 1952-1955

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INTRODUCTION

In this supplement are listed 111 diseases of fungous, bacterial, virus, and physiological origin, on 162 host plants. The list includes diseases and host plants recorded in publications since the preparation of the "First Supplement" (Brien and Dingley 1953), new records, and additional host records.

Among the fungous diseases listed the following are of special economic importance: *Phytophthora erythroseptica*, *Helminthosporium turicum*, *Monochaetia unicornis*, *Urophlyctis alfalfae*, and *Puccinia antirrhini*. *Verticillium dahliae* is listed on nineteen and *Pseudomonas syringae* on eighteen additional host plants. Eighteen new records of fungous diseases are included.

The virus diseases listed include cereal yellow dwarf, big-vein virus, cauliflower mosaic, citrus quick-decline, freesia mosaic, and lily mosaic, all of which are of particular economic importance.

No records of artificially infected host plants are given. An index of pathogens and physiological diseases is appended.

Abutilon sp.

Verticillium dahliae Kleb. Additional host record

Acacia pravissima F. V. Muell.

Verticillium albo-atrum Reinke & Berth. Additional host record

Acer pseudoplatanus L.

Verticillium dahliae Kleb. Additional host record

Actinidia chinensis Planch.

Armillaria mellea (Fr.) Quel. Additional host record

Aegeratum houstonianum Mill.

Verticillium dahliae Kleb. Additional host record

Antirrhinum majus L.

Puccinia antirrhini Diet. & Holw. Helson 1953

Arbutus sp.

Pseudomonas syringae van Hall Dye, D. W. and Dye, M. H. 1954.

Aster ericoides L.

Erysiphe cichoracearum DC. Additional host record

Aster laevis L.

Erysiphe cichoracearum DC. Additional host record

Aster novae-angliae L.

Erysiphe cichoracearum DC. Additional host record

Aster novi-belgii L.

Erysiphe cichoracearum DC. Additional host record

Avena sativa L.

Ophiobolus graminis Sacc. Additional host record
Cereal yellow dwarf Smith, H. C. 1955

Beilschmiedia tarairi (A. Cunn.) Benth. & Hook. f.

Glomerella cingulata (Stonem.) Spauld. & v. Schr. Additional host record

Betula alba L.		
<i>Pseudomonas syringae</i> van Hall		Dye, D. W. and Dye, M. H. 1954.
Boronia megastigma Nees		
<i>Verticillium dahliae</i> Kleb.		Additional host record
Brassica napo-brassica Mill.		
Cauliflower mosaic		Additional host record
Brassica rapa L.		
Cauliflower mosaic		Additional host record
Brassica oleracea L. var. botrytis L.		
Cauliflower mosaic		Fry 1952a
Brassica oleracea var. capitata L.		
<i>Alternaria brassicicola</i> (Schw.) Wilt.		New record
Cauliflower mosaic		Fry 1952a
Bromus catharticus Vahl.		
<i>Gibberella zeae</i> (Schw.) Petch		Additional host record
Bromus spp.		
Cereal yellow dwarf		Smith, H. C. 1955
Callistephus chinensis Nees		
<i>Erysiphe cichoracearum</i> DC.		Additional host record
Calystegia sepium R. Br.		
<i>Pseudomonas syringae</i> van Hall		Dye, D. W. and Dye, M. H. 1954
Camellia japonica L.		
<i>Exobasidium camelliae</i> Shirai		New record
<i>Sphaceloma</i> sp.		Additional host record
Chamaecyparis thyoides B.S.P.		
<i>Monochaetia uniconis</i> (Cke. & Ell.) Sacc.		Fuller and Newhook 1954
Chamaecyparis lawsoniana Parl.		
<i>Monochaetia uniconis</i> (Cke. & Ell.) Sacc.		Fuller and Newhook 1954
Chamaecyparis pisifera Sieb. & Zucc.		
<i>Monochaetia uniconis</i> (Cke. & Ell.) Sacc.		Fuller and Newhook 1954
Chrysanthemum morifolium Ramat		
<i>Verticillium dahliae</i> Kleb.		Additional host record
Citrullus vulgaris Schrad.		
<i>Verticillium dahliae</i> Kleb.		Additional host record
Citrus paradisi Macf.		
<i>Elsinoe fawcetti</i> Bitanc. & Jenkins		Additional host record
Citrus reticulata Blanco (Syn. <i>C. nobilis</i>)		
<i>Elsinoe fawcetti</i> Bitanc. & Jenkins		Additional host record
<i>Gibberella baccata</i> (Wallr.) Sacc.		Additional host record
<i>Sclerotinia sclerotiorum</i> (Lib.) de Bary		Additional host record
Citrus sinensis Osbeck		
Citrus quick decline		Chamberlain 1954
Citrus sp. (Citronelle)		
<i>Pseudomonas syringae</i> van Hall		Dye, D. W. and Dye, M. H. 1954.
Citrus sp. (New Zealand Grapefruit)		
<i>Pseudomonas syringae</i> van Hall		Dye, D. W. and Dye, M. H. 1954.
Clematis hexasepala DC.		
<i>Uredo puawhananga</i> Baylis		Baylis 1954
Clematis paniculata Gmelin		
<i>Puccinia alboclava</i> Baylis		Baylis 1954
<i>Uredo puawhananga</i> Baylis		Baylis 1954
Clematis vitalba L.		
<i>Uredo puawhananga</i> Baylis		Baylis 1954
Cryptostemma calandulacea R. Br.		
Tomato spotted-wilt		Chamberlain 1954
Cucurbita pepo L.		
<i>Botrytis cinerea</i> Pers.		Gillard 1951
<i>Pseudoperonospora cubensis</i> (Berk. & Curt.) Rostow.		Gillard 1951
<i>Erwinia carotovora</i> (Jones) Holland		Gillard 1951
Cupressus macrocarpa Hartw.		
<i>Monochaetia uniconis</i> (Cke. & Ell.) Sacc.		Fuller and Newhook 1954

Cydonia oblonga Mill.

- Glomerella cingulata* (Stonem.) Spauld. & v. Schr. Cunningham 1925
Verticillium dahliae Kleb. Additional host record
 Internal browning Bollard 1953e
 Manganese deficiency Atkinson and Bollard 1953

Cyphomandra betacea Sendt.

- Tree-tomato bootlace disease Chamberlain 1954

Cytisus scoparius Link

- Verticillium dahliae* Kleb. Additional host record

Dactylis glomerata L.

- Gibberella zeae* (Schw.) Petch Additional host record

Daphne odora Thunb.

- Pseudomonas syringae* van Hall Dye, D. W. and Dye, M. H. 1954

Daucus carota L. var. *sativa* DC.

- Botrytis cinerea* Pers. Additional host record
 Cucumber mosaic Chamberlain 1954

Delphinium cultorum Voss

- Pseudomonas delphinii* (E. F. Smith) Stapp Anon. 1952b

Dianthus caryophyllus L.

- Carnation mosaic Chamberlain 1954

Digitaria sanguinalis (L.) Scop.

- Gibberella zeae* (Schw.) Petch Additional host record

Diospyros kaki L. f.

- Botrytis cinerea* Pers. Farmer 1952
Pseudomonas syringae van Hall Farmer 1952

Euonymus japonicus L.

- Oidium* sp. Additional host record

Feijoa sellowiana Berg.

- Glomerella cingulata* (Stonem.) Spauld. & v. Schr. Additional host record

Ficus carica L.

- Glomerella cingulata* (Stonem.) Spauld. & v. Schr. Additional host record

Freesia refracta Klatt

- Freesia mosaic Chamberlain 1954

Fuchsia hybrida Voss

- Verticillium dahliae* Kleb. Additional host record

Garrya elliptica Douglas

- Septoria* sp. Helson 1952

Gazania rigens R. Br.

- Sclerotinia sclerotiorum* (Lib.) de Bary Additional host record

Gerbera jamesonii Bolus

- Phytophthora* sp. Cunningham 1952

Gladiolus sp. cult.

- Fusarium oxysporum* Schlecht. Ellis 1952

Hebe sp.

- Septoria exotica* Speg. Sydow 1924
 (syn. *Scoleciasis atkinsonii* Syd.)

Hirschfeldia incana (L.) Lagreze-Fossat

- Mycosphaerella brassicicola* (Duby) Oudem. Additional host record

Hordeum vulgare L.

- Cereal yellow dwarf Smith, H. C. 1955

Humulus lupulus L.

- Verticillium albo-atrum* Reinke & Berth. Helson 1954
 Hop virus (identity not ascertained) Chamberlain 1954

Hyoseyamus niger L.

- Tomato spotted-wilt Chamberlain 1954

Hypericum androsaemum L.

- Melampsora hypericorum* (DC.) Wint. Baker, 1955b

Hypericum perforatum L.

- Glomerella cingulata* (Stonem.) Spauld. & v. Schr. Additional host record

Iris xiphium L.					
<i>Didymellina macrospora</i> Kleb.					Additional host record
Jasminum primulinum Hemsl.					
<i>Botrytis cinerea</i> Pers.					Additional host record
Lactuca sativa L.					
Big vein virus ..					Cunningham 1952
Lagenaria patersonii Don					
<i>Verticillium dahliae</i> Kleb.					Additional host record
Lathyrus odoratus L.					
Pea streak virus					Chamberlain 1954
Lepidium ruderales L.					
<i>Albugo cruciferarum</i> S. F. Gray (syn. <i>Cystopus candidus</i>) (Pers. ex Chev.) Lev.					Baker, 1955a
Leptospermum scoparium Forst.					
<i>Capnodium walteri</i> Sacc.					Mulcock 1954
Libocedrus decurrens Torr.					
<i>Monochaetia unicornis</i> (Cke. & Ell.) Sacc.					Fuller and Newhook 1954
Lilium amabile Palbin					
Lily mosaic					Chamberlain 1954
Lilium auratum Lindl.					
<i>Botrytis elliptica</i> (Berk.) Cke.					Anon. 1953a
Lily mosaic					Chamberlain 1954
Lilium candidum L.					
Lily mosaic					Chamberlain 1954
Lilium chalcedonicum L.					
Lily mosaic					Chamberlain 1954
Lilium concolor Salisb.					
Lily mosaic					Chamberlain 1954
Lilium duchartrei Fr.					
Lily mosaic					Chamberlain 1954
Lilium formosanum Wallace					
Lily mosaic					Chamberlain 1954
Lilium henryi Baker					
Lily mosaic					Chamberlain 1954
Lilium japonicum Thunb.					
Lily mosaic					Chamberlain 1954
Lilium leucanthum Baker var. centifolium Stearn					
Lily mosaic					Chamberlain 1954
Lilium nobilissimum Makino					
Lily mosaic					Chamberlain 1954
Lilium paradalium Kellogg					
Lily mosaic					Chamberlain 1954
Lilium regale Wilson					
Lily mosaic					Chamberlain 1954
Lilium rubellum Baker					
Lily mosaic					Chamberlain 1954
Lilium sargentiae Wilson					
Lily mosaic					Chamberlain 1954
Lilium speciosum Thunb.					
Lily mosaic					Chamberlain 1954
Lilium sulphureum Baker					
Lily mosaic					Chamberlain 1954
Lilium testaceum Lindl.					
Lily mosaic					Chamberlain 1954
Lilium tigrinum Ker-Gawl.					
Lily mosaic					Chamberlain 1954
Lilium wardii Stap. ex Stern					
Lily mosaic					Chamberlain 1954
Linum usitatissimum L.					
<i>Ascochyta linicola</i> Naoum. & Vass.					Kerr 1953
Lobularia maritima Desv.					
<i>Peronospora parasitica</i> (Pers.) de Bary					Additional host record
Lonicera japonica Thunb.					
<i>Herpobasidium deformans</i> Gould					New record
Lonicera nitida Wils.					
<i>Verticillium dahliae</i> Kleb.					Additional host record

- Lupinus angustifolius** L.
Glomerella cingulata (Stonem.) Spauld. & v. Schr. Smith, H. C. 1955
- Lycopersicon esculentum** Mill.
Phytophthora parasitica Dastur Anon. 1953a
 Potato virus Y Chamberlain 1954
 Magnesium deficiency Johannesson 1951
- Matthiola incana** R. Br.
Erwinia carotovora (Jones) Holland Additional host record
- Medicago sativa** L.
Pellicularia filamentosa (Pat.) Rogers Additional host record
Stagonospora meliloti (Lasch) Petrak Smith, H. C. 1955
Urophlyctis alfalfae (Lagerh.) Magn. Smith, H. C. 1955
 Lucerne mosaic Fry 1952b
- Microlaena avenacea** Hook. f.
Claviceps purpurea (Fr.) Tul. Additional host record
- Microlaena stipoides** R. Br.
Claviceps purpurea (Fr.) Tul. Additional host record
- Morus alba** L.
Mycosphaerella alba (Fckl.) Lindau New record
- Narcissus** sp. cult.
Fusarium oxysporum Schlecht. Additional host record
- Nicotiana tabacum** L.
Erysiphe cichoracearum DC. Additional host record
Phytophthora parasitica Dastur Anon. 1952a
- Olea europaea** L.
Spilocaea oleaginea (Cast.) Hughes New record
- Osmanthus** sp.
Verticillium dahliae Kleb. Additional host record
- Passiflora edulis** Sims
Pseudomonas syringae van Hall Dye, D. W. and Dye, M. H. 1954
- Pelargonium domesticum** Bailey
Verticillium dahliae Kleb. Additional host record
- Pelargonium hortorum** Bailey
Verticillium dahliae Kleb. Additional host record
- Phaseolus vulgaris** L.
Fusarium solani Mart. var. *martii* Appel. & Wollenw. Brien *et al.* 1955
Sclerotium rolfsii Sacc. Additional host record
 Baldhead (physiological) Brien *et al.* 1955
 Blister pod (physiological) Brien *et al.* 1955
 Mottle leaf (physiological) Brien *et al.* 1955
 Sunscald (physiological) Brien *et al.* 1955
 Yellow spot (physiological) Brien *et al.* 1955
- Phebalium billardieri** Juss.
Phytophthora cactorum (Leb. & Cohn) Schroet. Anon 1953b
Verticillium dahliae Kleb. Anon 1953b
- Phlox paniculata** L.
Cercospora omphacodes Ellis & Holway New record
Septoria phlogis Sacc. & Speg. New record
Verticillium dahliae Kleb. Additional host record
- Podalyria** sp.
Phytophthora sp. Cunningham 1952
- Polygonum persicaria** L.
Septoria polygonorum Desm. New record
- Populus nigra** L.
Pseudomonas syringae van Hall Dye, D. W. 1954b
- Prostranthera ovalifolia** R. Br.
Verticillium dahliae Kleb. Additional host record
- Prostranthera rotundifolia** R. Br.
Verticillium dahliae Kleb. Additional host record
- Protea compacta** R. Br.
Verticillium dahliae Kleb. Additional host record

- Prunus armeniaca** L.
Pseudomonas syringae van Hall Dye, D. W. 1954a
 Moorpark mottle Chamberlain, Atkinson, and
 Hunter 1954
 Manganese deficiency Bollard 1953a
- Prunus avium** L.
Fusicladium carpophilum (Thuem.) Oudem.
 (syn. *Cladosporium carpophilum* Thuem.) Additional host record
- Prunus communis** Fritsch
Pseudomonas syringae van Hall Dye, D. W. 1954a
- Prunus persica** Sieb. & Zucc.
Armillaria mellea (Fr.) Quel. Additional host record
Gibberella baccata (Wallr.) Sacc. Additional host record
Pseudomonas syringae van Hall Dye, D. W. 1954a
 Potash deficiency Bollard 1953d
 Zinc deficiency Bollard 1953c
- Prunus** spp. (Plums)
Pseudomonas syringae van Hall Dye, D. W. 1954a
 Hollow end (cause unknown) Farmer 1954
 Manganese deficiency Atkinson and Bollard 1953
- Pyrus angustifolia** Ait.
 Apple mosaic Chamberlain 1954
- Pyrus communis** L.
Neofabraea malicorticis (Cordl.) Jack Additional host record
Pythium sp. Atkinson 1953
Pseudomonas syringae van Hall Dye, D. W. and Dye, M. H.
 1954
 Black end (cause unknown) McKenzie 1951
 Core breakdown (physiological) Padfield 1954
 Pear scald (physiological) Padfield 1954
 Zinc deficiency Bollard 1953b
- Pyrus malus** L.
Pseudomonas syringae van Hall Dye, D. W. and Dye, M. H.
 1954
 Apple ring-spot (syn. Henderson spot) Atkinson, Chamberlain, and
 Hunter 1954
 Brown heart (physiological) Padfield 1954
 Calyx-end breakdown (cause unknown) Padfield and Woon 1954
 Core flush (physiological) Padfield 1954
 Gnarling (cause unknown) McKenzie 1953
 Lenticel spot (physiological) Padfield 1954
 Light-induced scald (physiological) Padfield 1954
 Manganese deficiency Atkinson and Bollard 1953
 Superficial scald (physiological) Padfield 1954
- Ribes sanguineum** Pursh
Verticillium dahliae Kleb. Additional host record
- Rorippa islandica** (Oeder) Borbas
Albugo cruciferarum S. F. Gray (syn.
Cystopus candidus (Pers. ex Chev.) Lev.) Baker, 1955a
- Rosa** sp. cult.
Sphaeloma rosarum (Pass.) Jenkins New record
Agrobacterium tumefaciens (Sm. & Townsd.)
 Conn. Additional host record
- Rhododendron** sp.
Pseudomonas syringae van Hall Dye, D. W. and Dye, M. H.
 1954
- Rubus cissoides** A. Cunn.
Sphaerotheca pannosa (Wallr.) Lev. Additional host record
- Rubus fruticosus** L.
Elsinoe veneta (Burk.) Jenkins Additional host record
- Rubus idaeus** L.
 Raspberry virus Cadman 1947
- Rubus phoenicolasius** Maxim.
Elsinoe veneta (Burk.) Jenkins Additional host record
- Salix fragilis** L.
Pseudomonas syringae van Hall Dye, D. W. 1954b

<i>Salix</i> sp.		
<i>Armillaria mellea</i> (Fr.) Quel.		Additional host record
<i>Sambucus nigra</i> L.		
<i>Cercospora depazeoides</i> (Desm.) Sacc.		New record
<i>Senecio cruentus</i> DC.		
<i>Alternaria senecionis</i> Neergaard		New record
Tomato spotted-wilt		Chamberlain 1954
<i>Shawia paniculata</i> Forst.		
<i>Verticillium dahliae</i> Kleb.		Additional host record
<i>Sherardia arvensis</i> L.		
<i>Peronospora calotheca</i> de Bary		New record
<i>Solanum aviculare</i> Forst. f.		
<i>Phytophthora infestans</i> (Mont.) de Bary		Additional host record
<i>Solanum tuberosum</i> L.		
<i>Phytophthora erythroseptica</i> Pethyb.		Burns 1953
Potato crinkle		Chamberlain 1954
<i>Spinacia oleracea</i> L.		
Cucumber mosaic		Chamberlain 1954
<i>Stellaria media</i> (L.) Vill.		
<i>Peronospora alsinearum</i> Casp. ex de Bary		Additional host record
<i>Syringa vulgaris</i> L.		
<i>Pseudomonas syringae</i> van Hall		Dye, D. W. 1954b
<i>Thuja occidentalis</i> L.		
<i>Monochaetia unicornis</i> (Cke. & Ell.) Sacc.		Fuller and Newhook 1954
<i>Thuja orientalis</i> L.		
<i>Pestalotia funerea</i> Desm.		Additional host record
<i>Trifolium dubium</i> Sibth.		
<i>Peronospora trifoliorum</i> de Bary		Additional host record
<i>Trifolium fragiferum</i> L.		
Lucerne mosaic		Fry 1952b
<i>Trifolium pratense</i> L.		
<i>Sclerotinia spermophila</i> Noble		Additional host record
Lucerne mosaic		Fry 1952b
<i>Trifolium repens</i> L.		
<i>Cercospora zebrina</i> Pass.		New record
<i>Sphaerulina trifolii</i> Rostr.		New record
<i>Triticum aestivum</i> L.		
Cereal yellow dwarf		Smith, H. C. 1955
<i>Veronica arvensis</i> L.		
<i>Peronospora grisea</i> (Ung.) de Bary		New record
<i>Viola odorata</i> L.		
Cucumber mosaic		Chamberlain 1954
<i>Viola tricolor</i> L. var. <i>hortensis</i> DC.		
<i>Cercospora violae</i> Sacc.		New record
<i>Vitis vinifera</i> L.		
<i>Pseudomonas syringae</i> van Hall		Dye, D. W. and Dye, M. H. 1954
<i>Zantedeschia</i> sp.		
Cucumber mosaic		Chamberlain 1954
<i>Zea mays</i> L.		
<i>Helminthosporium turcicum</i> Passer.		New record
<i>Gibberella zeae</i> (Schw.) Petch		Additional host record

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INDEX OF PATHOGENS AND PHYSIOLOGICAL DISEASES

Agrobacterium tumefaciens	32	Moorpark mottle	32
Albugo cruciferarum	30, 32	Mottle leaf	31
Alternaria brassicicola	28	Mycosphaerella alba	31
Alternaria senecionis	33	Mycosphaerella brassicicola	29
Apple mosaic	32	Neofabraea malicorticis	32
Apple ring-spot	32	Oidium sp.	29
Armillaria mellea	27, 32, 33	Ophiobolus graminis	27
Ascochyta linicola	30	Pea streak virus	30
Baldhead	31	Pear scald	32
Big vein virus	30	Pellicularia filamentosa	31
Black end	32	Peronospora alsinearum	33
Blister pod	31	Peronospora calotheca	33
Botrytis cinerea	28, 29, 30	Peronospora grisea	33
Botrytis elliptica	30	Peronospora parasitica	30
Brown heart	32	Peronospora trifoliorum	33
Calyx-end breakdown	32	Pestalotia funerea	33
Capnodium walteri	30	Phytophthora cactorum	31
Carnation mosaic	29	Phytophthora erythroseptica	33
Cauliflower mosaic	28	Phytophthora infestans	33
Cercospora depazeoides	33	Phytophthora parasitica	31
Cercospora omphacodes	31	Phytophthora sp.	29, 31
Cercospora violae	33	Potash deficiency	32
Cercospora zebrina	33	Potato crinkle	33
Cereal yellow dwarf	27, 28, 29, 33	Potato virus Y	31
Citrus quick-decline	28	Pseudomonas delphinii	29
Claviceps purpurea	31	Pseudomonas syringae	27, 28, 29, 31, 32, 33
Core breakdown	32	Pseudoperonospora cubensis	28
Core flush	32	Puccinia alboclava	28
Cucumber mosaic	29, 33	Puccinia antirrhini	27
Didymellina macrospora	30	Pythium sp.	32
Elsinoe fawcetti	28	Raspberry virus	32
Elsinoe veneta	32	Sclerotinia sclerotiorum	28, 29
Erwinia carotovora	28, 31	Sclerotinia spermophila	33
Erysiphe cichoracearum	27, 28, 31	Sclerotium rolfsii	31
Exobasidium camelliae	28	Septoria exotica	29
Freesia mosaic	29	Septoria phlogis	31
Fusarium oxysporum	29, 31	Septoria polygonorum	31
Fusarium solani var. <i>martii</i>	31	Septoria sp.	29
Fusicladium carpophilum	32	Sphaceloma rosarum	32
Gibberella baccata	28, 32	Sphaceloma sp.	28
Gibberella zeae	28, 29, 33	Sphaerotheca pannosa	32
Glomerella cingulata	27, 29, 31	Sphaerulina trifolii	33
Gnarling	32	Spilocaea oleaginea	31
Helminthosporium turcicum	33	Stagonospora meliloti	31
Herpobasidium deformans	30	Sunsald	30, 31
Hollow end	32	Superficial scald	32
Hop virus	29	Tomato spotted-wilt	28, 29, 33
Internal browning	29	Tree tomato bootlace disease	29
Lenticel spot	32	Uredo puawhananga	28
Light-induced scald	32	Urophlyctis alfalfae	31
Lily mosaic	30	Verticillium albo-atrum	27, 29
Lucerne mosaic	31, 33	Verticillium dahliae	27, 28, 29, 30, 31, 32
Magnesium deficiency	31	Yellow spot	31
Manganese deficiency	29, 32	Zinc deficiency	32
Melampsora hypericorum	29		
Monochaetia unicornis	28, 30, 33		

METHODS FOR DETERMINING THE VIABILITY OF VARIOUS SEEDS BY TETRAZOLIUM STAINING

III. ITALIAN RYEGRASS AND SHORT-ROTATION RYEGRASS

(Received for publication, 24 March 1955)

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Summary

The viability of seed of Italian ryegrass and short-rotation ryegrass was determined by tetrazolium staining within 24 hours. Seeds were presoaked in water for 16 hours at 30°C. and surface sterilized with calcium hypochlorite. The embryos were then bisected and the seeds steeped in a 1% solution of 2, 3, 5-triphenyltetrazolium bromide for 6 hours at 30°C. Seeds were regarded as viable if all the embryo apart from the coleorhiza and the root primordium became stained.

With fully after-ripened seed, comparative tests by the germination and staining methods were in close agreement.

After-ripening of newly harvested seed stored at room temperature and 60% relative humidity occupied 8 to 12 weeks, and in viability tests made during this period dormant seeds reacted to the stain.

It is concluded that the staining method of determining the viability of seed of Italian ryegrass and short-rotation ryegrass is satisfactory and is more reliable than the germination method for testing seed during the after-ripening period.

INTRODUCTION

Experience in seed testing indicated that the seeds of Italian ryegrass *Lolium multiflorum* Lam. and of the hybrid strain "short-rotation ryegrass" tend to be dormant when newly harvested. Because of this dormancy, a reliable determination of the proportion of viable seeds in newly harvested samples cannot readily be made by the germination method. Furthermore, it is impracticable to postpone germination tests until after-ripening is complete because, in New Zealand, much of the seed harvested in January is required for sowing in March of the same year. There is need for a method of determining the viability of these seeds that is not subject to errors due to dormancy.

It has already shown that tetrazolium staining reliably indicates viability in seeds of numerous grasses, including perennial ryegrass (Hyde 1952). Furthermore, Porter *et al.* (1947) have shown in their work with *Paspalum notatum* that dormant seeds may react to the stain. The present study was undertaken to ascertain whether the staining method could be used for determining the viability of seed of Italian ryegrass and short-rotation ryegrass, especially seed in the dormant condition.

MATERIAL

For comparative viability tests, commercial samples of certified seed were used, 12 samples of Italian ryegrass and 12 of short-rotation ryegrass. The age of the seed ranged from 10 months to 3 years.

The observations on dormancy were made on 2 samples of pedigree seed harvested at Palmerston North in January 1952 and 2 samples harvested in January 1953.

METHODS

Staining tests for viability were carried out on samples of 400 seeds. The method was the same as that used with perennial ryegrass (Hyde 1952). The seeds were pre-soaked in water for 16 hours at 30°C., disinfected with calcium hypochlorite, bisected through the embryo and lower half of the endosperm, and steeped in a 1% solution of 2, 3, 5-triphenyltetrazolium bromide for 6 hours at 30°C. Seeds were counted as viable if all the embryo apart from the coleorhiza and root primordium became stained. Germination tests were made with samples of 400 seeds incubated for 14 days on moist filter paper with exposure to diffused daylight. Night and day temperatures were 18°C. and 25°C. respectively. A seed was counted as viable if it produced a plumule and one or more seminal roots.

The following series of tests were carried out:

1. Comparative tests on the fully after-ripened commercial samples.
2. Comparative tests on newly harvested seed stored at room temperature and at a relative humidity of approximately 60%. These tests were repeated at intervals of 14 days from the time of harvesting until after-ripening was complete.
3. Special germination tests prolonged for 56 days were carried out on the newly harvested seed and repeated, with 14-day intervals between the dates of commencement, until after-ripening was complete.

TABLE 1. RESULTS OF COMPARATIVE VIABILITY TESTS ON 24 SAMPLES OF FULLY AFTER-RIPENED SEED BY THE STAINING AND GERMINATION METHODS

Description of Sample	Viable Seed Per Cent	
	Germination test	Staining test
Italian ryegrass	41.7	44.2
" "	96.2	97.7
" "	94.5	94.2
" "	90.5	92.2
" "	94.7	96.7
" "	82.2	78.2
" "	91.5	88.7
" "	96.7	98.0
" "	83.0	81.5
" "	86.0	91.2
" "	92.0	95.2
" "	56.2	56.7
Short-rotation ryegrass	92.2	92.0
" "	96.5	94.5
" "	82.2	82.2
" "	88.7	91.5
" "	66.7	66.2
" "	78.5	76.2
" "	79.7	82.5
" "	72.2	71.0
" "	91.7	95.0
" "	74.5	75.2
" "	90.5	90.0
" "	72.0	75.2

RESULTS

The percentages of viable seeds determined in comparative tests on the commercial samples are shown in Table 1.

The regressions of germination percent (y) on staining percentage (x) were as follows:

$$\text{Italian ryegrass } y = +0.3773 + 0.9862x$$

$$\text{Standard error of the regression coefficient } \pm 0.0473$$

$$\text{Short-rotation ryegrass } y = +2.7455 + 0.9607x$$

$$\text{Standard error of the regression coefficient } \pm 0.0637$$

The results of successive comparative tests of newly harvested seed stored in the laboratory are shown in Table 2. Results of successive prolonged germination tests are presented in Fig. 1. Results from only one typical sample are shown.

TABLE 2. PERCENTAGES OF VIABLE SEEDS FROM COMPARATIVE TESTS BY THE GERMINATION AND STAINING METHODS MADE SUCCESSIVELY ON NEWLY HARVESTED SEED

Time in weeks from harvest to commencement of test	Italian Ryegrass				Short-rotation Ryegrass			
	Harvested 1952		Harvested 1953		Harvested 1952		Harvested 1953	
	Germ.	Stain.	Germ.	Stain.	Germ.	Stain.	Germ.	Stain.
0	49	95	60	95	76	98	47	96
2	63	93	65	93	87	97	53	94
4	81	96	76	97	87	97	64	94
6	86	92	79	96	90	97	68	95
8	94	95	88	95	94	98	84	94
10	93	94	89	94	95	98	87	96
12	95	96	94	95	97	97	93	93
14	94	95	95	94	97	98	95	95

DISCUSSION

Comparative tests on fully after-ripened seeds show that the staining method gave a measure of viability in close agreement with the germination tests (Table 1). The regressions of staining on germination did not differ significantly from $y = x$.

The newly harvested seed of Italian ryegrass and short-rotation ryegrass contained from 20% to 50% of dormant seed, i.e., viable seed which failed to germinate within the 14-day period of the standard germination test. After-ripening was complete after 2 to 3 months storage during which the seed increased gradually in germinating capacity. Throughout this period, germination tests failed to give a true measure of the percentage of viable seed: even when incubated

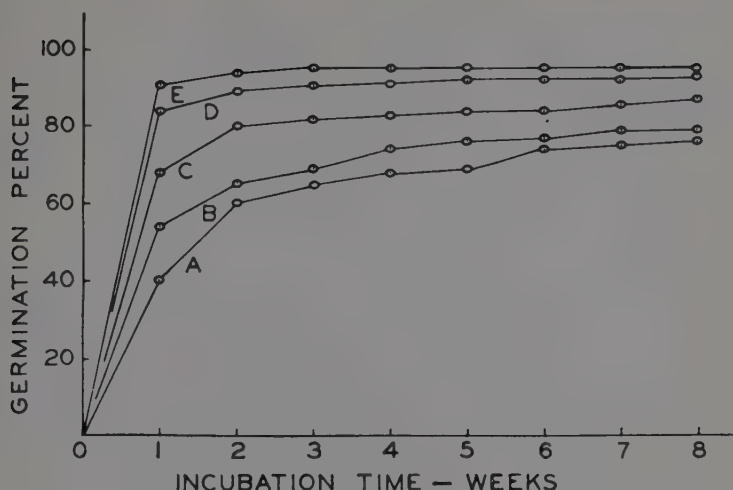


Fig. 1.—Cumulative germination percentages from prolonged tests made successively on Italian ryegrass seed harvested in January 1953. Interval between harvest and commencement of test: A 0 weeks, B 2 weeks, C 4 weeks, D 8 weeks, E 12 weeks.

for 56 days some of the viable seed failed to germinate. On the other hand, tetrazolium staining gave at all stages an accurate measure of the percentage of viable seeds and indicated what the germinating capacity determined by the standard 14-day test would be when after-ripening was complete.

The prolonged germination tests on newly harvested seed (Fig. 1) indicate that the factor inhibiting germination diminished slightly faster under storage conditions than under germinating conditions. Prolonging the germination test was not an efficient method of determining the viability of new seed.

It is concluded that with fully after-ripened seed of Italian ryegrass and short-rotation ryegrass the tetrazolium staining method is equally as reliable as the germination method for determining the germinating capacity, and that during the three months following harvesting tetrazolium staining gives a more accurate measure of the value of these seeds than does the germination test.

ACKNOWLEDGEMENT

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EFFECT OF PHOTOPERIOD ON VARIETIES OF RED CLOVER (*Trifolium pratense* L.)

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Summary

Two varieties of red clover have been grown under photoperiods ranging from 12 to 16 hours.

The varieties reacted differently to changes in photoperiod.

For the purpose of distinguishing between the two varieties, leaf size, length of petiole, and time of commencing flowering, were the most useful characters.

Differences in length of petiole and in length of flowering stem were most significant in a 12-hr. photoperiod.

Differences in flowering were highly significant in 13-, 13½-, and 14-hr. photoperiods.

Differences in size of leaflet were independent of photoperiod.

A 13-hr. photoperiod was most favourable for tiller development in the broad red clover as against a 14-hr. period for the late-flowering variety.

The varieties differed in number of internodes but gave no photoperiod $\times$ variety interaction.

Chilling treatments had no useful effects and resulted only in a depression of growth.

INTRODUCTION

In New Zealand, the two varieties of red clover (*Trifolium pratense* L.) used in pasture mixtures are the early flowering broad red, and the late-flowering Montgomery. Both of these are included in the official seed-certification scheme, which is operated on a system of field inspection, followed by plot trials as a check test on lines certified.

In the plot trials, which require a full year before a final report can be given, the two varieties are distinguishable by their differences in growth habit, and in growth rate in response to the lengthening days and rising temperatures of the early spring months. A test which would differentiate between these and other varieties within a relatively short time is desirable. The object of the experiments described below was to study the photoperiodic behaviour of two varieties of red clover, to see if under any particular set of conditions they would react differently, and thereby provide the basis for a short test which might replace the present certification procedure.

Since Garner and Allard (1920) published their results on the effects of relative lengths of day and night on growth and reproduction of plants, many experiments have been conducted on the effects of photoperiodism on plant growth. Various herbage plants have been studied and classified as long-day or short-day plants (Garner and Allard 1923; Evans and Allard 1934; Allard and Evans 1941). Red clover has been included in some of these experiments. Pohjakallio (1951) found that in a 10-hr. photoperiod red clover was dwarfed in growth compared with the normal growth in a day of 18 hr. in south Finland.

Keller and Peterson (1950) included two varieties of red clover in a pot trial where the clovers were grown individually, and in association with different strains of timothy (*Phleum pratense* L.). In their experiments no flowers developed on red clover plants subjected to a 10-hr. day whereas with an 18-hr. day there were numerous flowers.

The effect of light on the growth and development of the Dollard variety of red clover has been studied by Ludwig, Barrales, and Steppler (1953). Each of their clonal lines flowered in a photoperiod of 16 hr.; all but two in 14 hr.; and none in 12 hr. or less.

Some pasture plants require exposure to low temperatures before they are able to respond to long photoperiods. Cooper (1952) found that in plants of perennial ryegrass (*Lolium perenne*) an overwintering exposure to low temperatures was required, and the late-flowering strains required a longer period of low temperatures than did the earlier-flowering strains. Similarly the late-flowering Tallarook variety of subterranean clover (*Trifolium subterraneum*) has been found by Aitken (1951) to require a period of low temperature before responding to long photoperiods. No reference was made to the effect of low temperatures on the development of red clover.

EXPERIMENTAL TECHNIQUE

Greenwood (1950) has shown that, in subterranean clover, seedling development is dependent on seed size, so for this experiment seeds of a uniform size were drawn from bulk samples of the New Zealand certified cowgrass (broad red clover) and from the Welsh Plant Breeding Station's S.123 strain of late-flowering red clover. These strains were taken as being representative of the two red clovers harvested and certified in New Zealand (Gorman 1949).

The seed samples were placed in a germinator for two days; germinating seedlings were then transferred from the germinator pads to 8-in. unglazed pots filled with a steam-sterilized potting soil, to which artificial fertilizers had been added. Four seedlings, later reduced to two, were planted in each pot. Each plant was identifiable at all stages of the experiment. All pots were inoculated with a culture of *Rhizobium* prepared from vigorous red clover plants. The pots were then placed in trays, and kept in a glasshouse with a minimum night temperature of 45° to 50°F.

For the first experiment the layout was a 5 x 2 x 4 factorial design with only one replication, but with four plants in each subclass (i.e., two plants in each of two pots). Pots within groups, and groups within the glasshouse, were frequently re-randomized to eliminate variation other than that due to treatments.

The first chilling treatment was applied to 45-day-old seedlings. The treatment consisted of holding batches of plants for 6 days in a low-temperature chamber running at 36° to 38°F. Limitations of space prevented the chilling of all plants simultaneously.

Light treatments were commenced on the 65th day after sowing, as soon as the chilling treatments were completed. Lighting consisted of daylight supplemented by artificial light from 200-watt bulbs placed four feet above the plants, one light to each group of plants. The

A typical layout of pots in each group was as follows:—

GROUP 1

Bd	Mb	Md	Bc
Mc	Ma	Ma	Mb
Mc	Bd	Bb	Ba
Bd	Ba	Bc	Md

B = N.Z. certified cowgrass
(broad red clover)

M = Welsh P.B.S. Strain S.123
(late-flowering type)

a = not chilled

b = 6 days chilling in early seedling
stage

c = chilling one week later than b

d = chilling one week later than c

Total number of pots = 80

Total number of plants = 160

Group 1 12 hr. light, 12 noon—
midnight

Group 2 13 hr. light, 11 a.m.—
midnight

Group 3 14 hr. light, 10 a.m.—
midnight

Group 4 15 hr. light, 9 a.m.—
midnight

Group 5 16 hr. light, 8 a.m.—
midnight



Fig 1.—Arrangement of pots in light boxes.

artificial lighting gave a light intensity of 5 foot-candles at ground level as against the minimum recommendation of 2 f.c. (Post 1942). Each group of plants in the glasshouse stood in a light-tight box fitted with a removable hinged lid (Fig. 1).

Each afternoon the lids were balanced in an upright position, and held by a wire loop which was part of the electrical circuit. A time clock switched on the lights an hour before sunset, and at midnight closed a circuit which extinguished the lights, broke the wire loop, and released the box lids. The following morning at 8 a.m. the lid of the 16-hr. box was removed and the remaining lids were removed at hourly intervals. This was repeated daily for four weeks, discontinued for two weeks to reduce the spread of *Sclerotinia* infection, and resumed again for a final period of nine weeks. The first experiment was in progress from July 1951 until mid January 1952.

A second experiment using a similar number of plants and a similar arrangement was commenced in March 1952 and completed in February 1953. Controlled daylengths in this experiment were 12 hr., 12½ hr., 13 hr., 13½ hr., and 14 hr. and no chilling treatments were applied.

TABLE 1. LENGTH OF LONGEST STEM, WIDTH AND LENGTH OF CENTRE LEAFLET AT 64 DAYS, AND WEIGHT OF CUTTINGS AT 74 DAYS
Combined Analyses of Variance

Source of Variation	d.f.	F Required 5% (1%) points	Mean Squares and Results of F Tests †			
			Stem Length (mm.) 64 days	Leaflet Width (mm.) 64 days	Leaflet Length (mm.) 64 days	Weight (g.) 74 days
Group	4					
Variety	1	4.20 (7.64)	14,803.25**	302.50**	823.56**	13.163**
Chilling	3	2.95 (4.57)	2,707.72**	57.13**	135.39**	10.498**
V x C	3		638.72	9.57	11.31	0.505
Residual	28		361.10	9.72	14.99	0.869
Individual plants	120		399.68	7.67	9.62	0.491
Varietal means and S. E.		Broad	105.4 ± 2.12	19.62 ± 0.35	23.22 ± 0.43	2.04 ± 0.104
		Late flowering	86.2	16.86	18.68	1.46
Coefficients of variation (per plant)			20.9%	15.2%	14.8%	40.1%

† In all the statistical tests in this paper, the following symbols are used:

*** Differences significant at the 0.1% level of probability.

** Differences significant at the 1% level of probability.

* Differences significant at the 5% level of probability.

RESULTS

First Experiment

Measurements of leaf size and stem length were taken on the sixty-fourth day after germination of the seedlings. This was on completion of the chilling treatments, and just prior to application of the light treatments. Ten days later, in order to control excessively tall growth, all plants were cut back. The cuttings from each plant were weighed.

An analysis of these weights and the measurements taken previously are combined in Table 1.

At this stage the broad red variety had the longer stems, the larger leaflets, and the greater yield of fresh herbage. The chilling treatments had caused a significant retardation of growth, but this had been no greater in one variety than in the other, as shown by the lack of significant interactions.

On the 70th day after commencing light treatments (60 days from cutting back) the height of each plant was measured, and the number of tillers on each plant counted. An analysis of these is given in Table 2.

TABLE 2. EFFECT OF PHOTOPERIOD ON PLANT HEIGHT AND NUMBER OF TILLERS
Analyses of Variance

Source of Variation	d.f.	F Required 5% (1%)	Mean Squares and Results of F Tests	
			Plant height (in.)	No. of tillers ($\sqrt{1 + X}$)
Photoperiod	4	2.46 (3.51)	346.71**	3.993**
Variety	1	3.94 (6.90)	577.60**	0.014
Chilling	3	2.70 (3.98)	6.69	0.099
P x V	4	2.46 (3.51)	33.81	0.350*
P x C	12	1.85 (2.36)	24.86	0.090
V x C	3	2.70 (3.98)	15.53	0.252
P x V x C	12	1.85 (2.36)	6.63	0.061
Residual	102		23.84	0.109
Coefficients of variation			17.6%	22.4%

The means of plant height for the 12-hr. and 13-hr. photoperiods were significantly lower than those for the 11-, 15-, or 16-hr. groups. The broad red variety was significantly taller than the late flowering, the varietal means and their standard error being 9.78 ± 0.028 and 7.88 ± 0.028 in. respectively.

There was no photoperiod $\times$ variety interaction for height at this stage of growth.

For number of tillers the variety means did not differ significantly but there was a significant photoperiod $\times$ variety interaction.

The table of means (transformed) and the differences required for significance are:—

	12 hr.	13 hr.	14 hr.	15 hr.	16 hr.	Variety Means
Broad red	1.03	1.30	1.54	1.68	1.78	1.47
Late flowering	1.04	1.00	1.81	1.80	1.77	1.48

Significant difference at 5%: Variety means—0.103

Variety means within photoperiods—0.231

In the 12-, 15-, and 16-hr. groups the varieties did not differ significantly. In the 13-hr. group the mean of the broad red was significantly greater than the mean of the late flowering strain, whereas in the 14-hr. group the position was reversed, the mean of the late flowering being significantly the greater.

This indicated that, in this experiment, as daylength was increased to 13 and 14 hr. the broad red responded at 13 hr., but the late flowering type did not respond till 14 hr., at which day-length it passed the broad red in tiller count, and then practically ceased to react to increasing day lengths (Fig. 2).

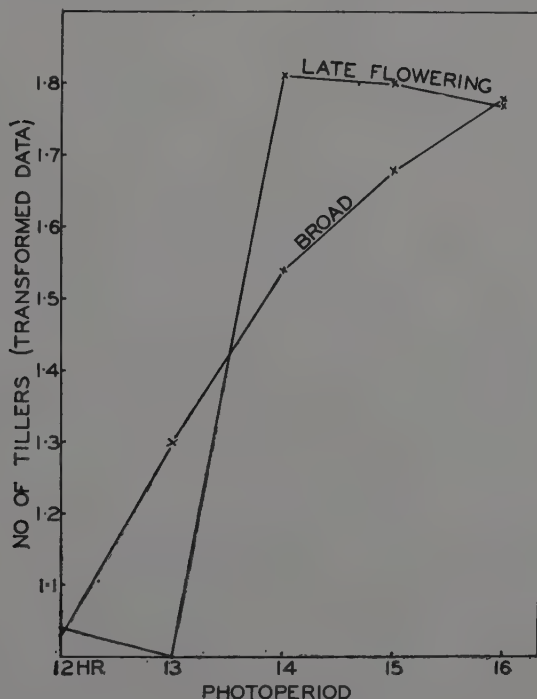


Fig. 2.—Effect of photoperiod on tiller formation.

The first flower buds emerged on the 112th day from sowing. A plant was considered to have commenced flowering when the first florets on the first bud were open for fertilization. Every third day,

for a period of ten weeks, a record was taken of plants that had commenced flowering. Prior to flowering some plants became infected with crown rot (*Sclerotinia trifoliorum* Erikss.). The incidence of this disease was most severe in the groups that received least daylight, and a number of infected plants died before reaching the flowering stage. During the flowering period there were a number of abortive inflorescences, particularly in the groups with the shortest day length. The flowering data are given in Table 3.

TABLE 3. NUMBERS OF PLANTS FLOWERING 192 DAYS AFTER SOWING

Photoperiod	Variety	Flowered	Not Flowered	X ₀
12 hr.	Broad	4	5	1.91*
	Late-flowering	0	11	
13 hr.	Broad	15	1	3.794**
	Late-flowering	3	12	
14 hr.	Broad	11	3	—
	Late-flowering	11	2	
15 hr.	Broad	15	1	—
	Late-flowering	13	3	
16 hr.	Broad	16	0	—
	Late-flowering	15	1	

There was a highly significant interaction between variety and photoperiod. In the 13-hr. photoperiod, 15 out of the possible 16 broad red plants had flowered as against 3 only of the late-flowering variety. There was a significant interaction in the 12-hr. photoperiod also, even though a number of plants had died before flowering commenced.

The results from the above experiment indicate that the greatest differences in growth and flowering were in the 12-hr. to 14-hr. range, where interactions for tillering and flowering were obtained.

Second Experiment

In the 1952-53 experiment, the treatments covered the 12- to 14-hr. photoperiods in half-hourly intervals. Since in the first experiment there had been no interactions between chilling treatments and variety, chilling treatments were omitted. No measurements were taken until after 33 days of controlled lighting. This was 130 days from the time of sowing. The leaf dimensions and length of petiole were then measured, and the number of tillers at the crown of each plant counted. An analysis of these is given in Table 4.

For both characters the mean for the 12-hr. group was significantly lower than that for any other photoperiod. There was otherwise little difference between the remaining groups except that for length of leaflet the mean for the 14-hr. photoperiod was significantly higher than the means for the 12½-hr. and 13-hr. photoperiods.

TABLE 4. ANALYSES OF VARIANCE ON MEASUREMENTS 130 DAYS AFTER SOWING

Source of Variation	d.f.	Mean Squares and Results of F Tests				
		F Reqd. 5% (1%)	Leaflet width (mm.)	Leaflet length (mm.)	Petiole length (mm.)	Number of tillers converted to $\sqrt{x + .5}$
Photoperiod	4	2.43 (3.44)	36.397*	152.39**	5,652.68**	0.6098**
Variety	1	3.91 (6.81)	529.256**	1543.81***	28,116.51**	2.3766**
P x V	4	2.43 (3.44)	2.1469	16.07	1,755.49*	0.2890*
Residual	148		11.10	21.94	708.35	0.0867
Coefficient of variation			13.7%	16.9%	13.3%	11.0%

The group means for leaflet width and leaflet length were as follows:

Photoperiod	12 hr.	12½ hr.	13 hr.	13½ hr.	14 hr.	Sig. diff. 5% (1%)
Leaflet width (mm.)	22.5	25.2	24.2	24.6	25.0	1.65 (2.17)
Leaflet length (mm.)	24.4	28.0	27.5	28.7	30.3	2.31 (3.06)

The varieties differed at the 1% level of probability in both width and length of leaflet, the means for the width being 26.15 mm. and 22.50 mm. for the broad red and late flowering respectively and the lengths 30.90 mm. and 24.70 mm. There was no evidence of variety $\times$ photoperiod interaction for size of leaflet.

The broad red had the longer petiole, and there was a significant variety $\times$ photoperiod interaction for this character.

The table of means for petiole length is as follows:

	12 hr.	12½ hr.	13 hr.	13½ hr.	14 hr.	Variety Means
Broad	203.5	212.5	208.0	210.5	224.5	211.8
Late flowering	153.0	181.0	193.0	194.0	205.5	185.4

Significant difference at 5% (1%): Variety means—8.3 (10.9)

Variety means within photoperiods—18.5 (24.4)

In the 12-hour photoperiod, the mean length of petiole in the broad red variety was significantly greater at the 0.1% level, and in the 12½-hr. photoperiod at the 1% level. In the remaining groups the differences failed to reach significance, except in the 14-hr. photoperiod where it was significant at the 5% level only. It is of interest to note that the greatest differences in leaflet size and petiole length occurred in the group with the shortest photoperiod.

The table of means for number of tillers (converted to $\sqrt{x + .5}$) is as follows:

	12 hr.	12½ hr.	13 hr.	13½ hr.	14 hr.	Variety Means
Broad	2.65	2.58	2.41	2.62	2.51	2.556
Late flowering	2.75	2.70	2.69	3.18	2.68	2.800

Significant difference at 5% (1%):

Variety means—0.092 (0.122)

Variety means within photoperiods—0.206 (0.272)

The greatest difference in means occurred in the 13½-hr. group when the mean number of tillers in the late-flowering variety was highly significantly the greater. The only other significant difference was in the varietal means of the 13-hr. group.

The first flower buds appeared on plants in the 14-hr. photoperiod 168 days from the time of sowing and 76 days after the adjustment of daylength. Just prior to the commencement of flowering, records were made of length of flowering stems, and of number of internodes.

An analysis of the lengths of flowering stems (transformed to log. cm.) and of the number of internodes is given below in Table 5.

TABLE 5. EFFECT OF PHOTOPERIOD ON GROWTH CHARACTERS PRIOR TO THE COMMENCEMENT OF FLOWERING (170 DAYS AFTER SOWING)

Source of Variation	d.f.	F Required 5% (1%)	Mean Squares and Results of F Tests	
			Length of longest flowering stem (log. cm.)	No. of internodes
Photoperiod	4	2.43 (3.44)	1.035**	6.088**
Variety	1	3.91 (6.81)	12.635***	15.006**
P x V	4	2.43 (3.44)	0.253*	0.819
Error	148		0.074	1.551
Coefficient of variation			21.0%	17.3%

For length of flowering stem there was a significant interaction of photoperiod x variety in that the differences between the varietal

means became significantly more pronounced with change of photoperiod from 12-hr. to 14-hr. This is shown graphically in Fig. 3. The ratio broad: late-flowering decreased progressively from 5.1 : 1 in the 12-hr. group to 2.3 : 1 in the 14-hr. group.

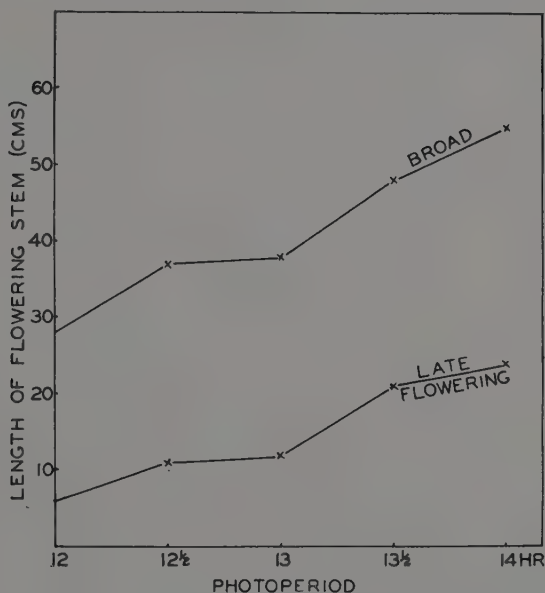


Fig. 3.—Effect of photoperiod on length of flowering stem.

The means for the two varieties differed at the 1% level of probability, the true means being broad 41.4 cm. and the late-flowering 14.6 cm.

In number of internodes the photoperiod means were:

12 hr.	12½ hr.	13 hr.	13½ hr.	14 hr.	Sig. diff. 5%
6.68	7.18	6.88	7.68	7.60	0.615

The 13½ and 14-hr. groups therefore had significantly higher means than the 12- or 13-hr. groups. The means for varieties differed at the 1% level these being 7.51 for the broad-red and 6.90 for the late-flowering variety, but changing photoperiod did not alter these differences.

Flower buds were emerging from the first week in September until late in January of the following year. An analysis of the subsequent flowering data, with a logarithmic transformation to stabilize variances, and the table of means are as follows:

TABLE 6. EFFECT OF PHOTOPERIOD ON FLOWERING
Analyses of Variance log (days after 30/8/52)

Source of Variation	d.f.	Mean Sq.	F	5% (1%) points of F
Photoperiod	4	2.7169	37.2**	2.44 (3.47)
Variety	1	21.0300	238.1**	3.92 (6.84)
P x V	4	0.7018	9.62**	2.44 (3.47)
Residual	131	0.0729		

Coefficient of Variation 16.3%

Table of Means
(transformed data)

	12 hr.	12½ hr.	13 hr.	13½ hr.	14 hr.	Variety Means
Broad	1.74	1.60	1.47	0.93	0.75	1.30
Late flowering	2.15	2.13	2.09	1.95	1.80	2.02

Significant difference at 5% (1%):

Variety means - - - - - 0.084 (0.112)

Variety means within photoperiods - - - - 0.189 (0.250)

The significant interaction in this analysis showed that the difference in flowering date (on a log scale) of varieties within photoperiod groups was significantly greater in the 13½- and 14-hr. groups than it was in any of the other groups. The ratio Late-flowering: Broad red, in days to flower from a base date, decreased significantly from 11.2 in the 14-hr. photoperiod to 2.58 in the 12-hr. photoperiod.

TABLE 7. NUMBER OF PLANTS FLOWERING ON 197TH DAY

	Variety	Flowered	Not Flowered	χ^2
14 hr.	B.	16	0	4.91**
	L. F.	0	12	
13½ hr.	B.	10	0	4.72**
	L. F.	0	16	
13 hr.	B.	10	5	3.49**
	L. F.	0	15	
12½ hr.	B.	5	7	2.10*
	L. F.	0	13	
12 hr.	B.	3	13	1.22
	L. F.	0	16	

In Fig. 4 this shows as a decreasing linear distance between the corresponding points in the graph for the two varieties.

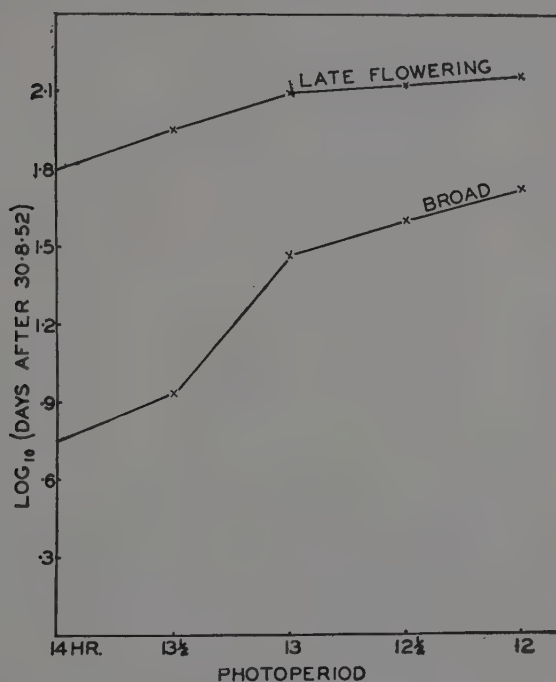


Fig. 4.—Effect of photoperiod on flowering date.

The mean flowering-date of the broad red was significantly earlier than that for the late-flowering, the difference averaging 85 days.

Although flowering extended over a period of four months, there were clearly defined differences between groups in rapidity of flowering, and in completion of flowering. This is illustrated by Tables 7 and 8, which were compiled from data early in the flowering season, and again three months later, to demonstrate the changes in that period.

There were highly significant differences in the 13-, 13½- and 14-hr. photoperiods. In both the 13½- and 14-hr. photoperiods there were no late-flowering plants in bud until after all the broad-red plants had commenced flowering.

At this later date, significant differences had been attained in the 12-hr., 12½-hr., and 13-hr. photoperiods, but the differences were not as clearly defined as at the earlier date.

Under the longer photoperiods of 13½ and 14 hr., plants of both varieties were stimulated towards flower-bud formation and extrusion of florets. The response of the two varieties to the longer photoperiods was quite different, so that within a few weeks of the

TABLE 8. NUMBERS OF PLANTS FLOWERING ON 287TH DAY

	Variety	Flowered	Not Flowered	χ_e
14 hr.	B.	16	0	—
	L. F.	12	0	
13½ hr.	B.	10	0	1.16
	L. F.	12	4	
13 hr.	B.	14	1	3.03**
	L. F.	5	10	
12½ hr.	B.	12	0	3.83**
	L. F.	2	11	
12 hr.	B.	16	0	4.63**
	L. F.	2	14	

commencement of flowering a count of the flowering and non-flowering plants in either the 13½- or 14-hr. photoperiods would have served to identify unknown plants of either variety (Fig. 5).



Fig. 5.—Effect of 14-hr. photoperiod on Broad red clover (left) and Late-flowering red clover (right) at 190 days.

DISCUSSION

At each time of measurement there were significant differences between the varieties for each botanical character examined. Thus broad red had wider and longer leaflets, longer petioles and flowering stems, and greater height. It flowered earlier and was the first to complete flowering under all treatments.

Plants were tallest and development most rapid in the longest photoperiods. Crown rot caused the greatest number of deaths in the shortest photoperiod. Abortive inflorescences also occurred more frequently under these conditions. Generally, red clover plants of a similar age grown in the field were more robust and more disease-resistant than the plants in the experimental groups inside the glasshouse.

Some important interactions were apparent after the controlled daylengths had been operating for six weeks and longer. As was shown in the means for Table 2, in tiller development the broad red responded to the 13-hr. treatment, but the late-flowering variety required a 14-hr. day. In developmental growth characters such as length of petiole and length of flowering stem, the broad red made the greater response at the shortest daylengths, although there were still significant differences in the 14-hr. treatments. Flower-bud emergence and completion of flowering were most rapid under the longest day-lengths, and with a 13- or 14- hr. day the response from the broad red was significantly different from that of the late-flowering variety at the 1% level. At the end of the first four weeks after the commencement of flowering all the broad-red plants in the 13½-hr. and 14-hr. groups were flowering, but no flowers had appeared on the late-flowering plants.

There were significant differences in growth characters between the two varieties within the first 130 days after sowing. As the plants grew, the differences were more difficult to detect until the flowering stage was reached, when there was again a difference that could be defined and recorded. The two varieties could be distinguished readily by measurements of the size of leaflets, and this character was least affected by changes in photoperiod. Length of petiole could be used with confidence if the plants were grown under a 12-hr. or 12½-hr. photoperiod, and possibly tiller counts could be used under a 13½-hr. or 14-hr. photoperiod. The largest differences in length of flowering stem were obtained in the shorter photoperiods. Especially on the late-flowering plants the number of internodes on a stem was difficult to determine, and this measure was not reliable.

Flowering date gave a most reliable separation of varieties in a photoperiod range of 13- to 14-hr., but there were difficulties in growing the plants to this stage under artificial conditions. The height of the plants and their susceptibility to disease caused most concern.

In the two experiments there was a difference in time before flowering commenced, probably because the first trial was sown in July and the second in March of the following year, and there was not complete control of temperatures in either experiment.

The measurement of leaf size and petiole length of 130-day-old seedlings which had been under a 12-hr. photoperiod for at least 38 days would appear to be a practical test for seed certification purposes. Further work may show that differences could be determined in a much shorter time.

From the normal growth cycle of red clover in the field, it was reasonable to suppose that exposure to a cold period could be used to separate the two varieties. However, in these experiments the chilling treatments applied had little effect other than to retard the growth of all plants. No chilling $\times$ variety interactions were recorded. There

were some chilling $\times$ group interactions, but the effect of the chilling was to reduce the differences which were measurable in the unchilled groups.

ACKNOWLEDGEMENTS

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FERTILIZER, CULTIVATION, AND GRASSING TRIALS WITH PAMPAS GRASS (*Cortaderia selloana*)

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(Received for publication, 30 March 1955)

Summary

Pampas grass gave a significant response to the application of superphosphate plus sulphate of ammonia and sulphate of potash in the first year of the trial. In the second year there was also a response from superphosphate alone. Clean cultivation sufficient to control weed growth gave a higher yield than where grasses and clovers constituted the ground flora.

INTRODUCTION

When pampas grass (*Cortaderia selloana* (Schult.) Aschers. et Graebn.) was first introduced as a forage crop, there arose the question of its response to the application of fertilizers. Early trials, conducted without replication or the adoption of a careful weighing technique gave no indication that pampas responded to fertilizers. As a result, there are frequent statements in articles on pampas grass that no benefit is to be derived from their application. It would indeed be strange if this were true. At the same time, it has been claimed that the vigour of pampas growth is, in part at least, affected by the ground flora in the immediate vicinity of the plants. Some excellent stands have been noted where clovers formed a high proportion of the ground flora and poor stands where clovers were absent. McClean* has strongly advocated intercultivation as a means of increasing growth, and it was decided to try these cultural methods in this fertilizer trial.

EXPERIMENTAL PROCEDURE

A composite trial was laid down at Massey Agricultural College in the spring of 1949 to determine (a) the response to fertilizer application, (b) the effect of cultivation and ground flora on yield.

Owing to the unevenness of the establishment, the area was replanted in spring 1950. Clones from McClean's nursery were used as they give greater uniformity than seedlings.

Design of Plots

The design used for this trial, which was prepared by the Applied Mathematics Laboratory, Department of Scientific and Industrial Research, gave 4 replications of a 5 x 5 graeco-latin square. While this is a powerful design for minimizing the effect of soil variation, it does not allow a test to be made of the interaction of the two factors used, i.e. fertilizer and cultural treatments, so that they will be dealt with separately. In the first cut following the application of treatments, the F ratio obtained from the interaction when treated as a 5 x 5 factorial layout with 4 replications was only 0.65 for 16,72 d.f. There is thus no indication of any interaction between the two sets of factors, and the ordinary analysis, ignoring interaction, can be used with confidence.

* Mr. A. McClean first experimented with pampas grass as a forage crop in 1926 and subsequently established nurseries to supply plants to farmers.

Each plot contained 6 plants (3 x 2) at 7-ft. spacings, and each plot was separated from the next by a 7-ft. grass verge which was kept grazed by sheep.

Fertilizer Application

Fertilizers were applied to the plots on six occasions over a period of two years, commencing 24 October 1952. The following rates were applied per acre in each of the two years:

Control (C)

Superphosphate 4 cwt. (P)

Superphosphate 4 cwt. + sulphate of ammonia 3 cwt.* (P + N)

Superphosphate 4 cwt. + sulphate of potash 2 cwt. (P + K)

Superphosphate 4 cwt. + sulphate of potash 2 cwt. + sulphate of ammonia 3 cwt. (P + K + N)

The fertilizers were not evenly distributed over the plots but were concentrated in a radius of about 2 ft. around the plants.

Cultivation and Grassing Treatments

From the time of planting (Sept. 1950) until the second cut was made (Sept. 1952) the land between all of the plants was kept cultivated. Immediately after the second cut, the following cultivation and sowing treatments were superimposed on the plots.

No cultivation, no sown clover.

Cultivation only (sufficient to control weed growth).

Red clover.

White clover.

Red clover, white clover, perennial ryegrass.

The seeds germinated and plants established well, and during the first summer there was a marked differentiation in ground flora between the plots. Twelve months after sowing, however, difference in clover content between them was slight. The average clover content of the plots, estimated during October and December 1953, was as follows:

Area not sown	-	-	-	38½%
Cultivated	-	-	-	—
Red clover	-	-	-	47%
White clover	-	-	-	54½%
Grass and clover mixture	-	-	-	36½%

As a result of the trend towards uniformity of the different sowings no effects can be credited to them. In point of fact the statistical analysis at the end of the first year of treatment shows that no significance can be attached to the differential sowings. The comparison then revolved around clean cultivation as against grass and clover (both sown and volunteer) which was kept grazed by sheep.

* For every pound of sulphate of ammonia applied, 1½ lb. of ground limestone was included to neutralize acidity caused by the application.

Times of Cutting

Before the start of the trial, the plots were cut in February 1952 and again in September 1952; the weights of this second cut were used in the analysis of co-variance. Shortly afterwards, in October 1952, fertilizer treatments were commenced and grasses and clovers were sown.

The first experimental cut was made on 26 September 1953 and the second on 1 September 1954.

RESULTS

First Experimental Cut

The analysis of co-variance (Table 1) is derived from the green weight (lb.) per plot of six plants.

TABLE 1. ANALYSIS OF FIRST EXPERIMENTAL CUT
(Using pre-trial weights as concomitant data.)

Source of Variation	d.f.	Adjusted Mean Squares	F	5% (1%) points of F
Squares: rows and columns within squares	35			
Manurial treatments	4	1,530.5	3.88**	2.54 (3.68)
Cultural treatments	4	6,210.6	15.75**	2.54 (3.68)
Residual	55	394.3		
Total	98			

Coefficient of variation = 10.3%

**Significant at 1%.

There are highly significant differences between the adjusted fertilizer means.

There are also highly significant differences between the adjusted cultural treatment means.

The adjusted means (per plot) for the fertilizer treatments are:

C	-	-	-	182.0 lb.
P	-	-	-	184.5 lb.
P + N	-	-	-	200.5 lb.
P + K	-	-	-	196.5 lb.
P + K + N	-	-	-	199.8 lb.

The standard errors of the above means are approximately 4.4 and the differences required for significance at the 5% (1%) levels of probability are respectively 12.6 (16.8). (The S.E.s change slightly from treatment to treatment, and the differences required for significance change slightly with the means being compared, but these changes are so small that the figures quoted can be used for comparisons without serious objection.)

The fertilizer comparisons show the following:

The means for C and P do not differ significantly.

The means for P + N, P + K, and P + K + N do not differ significantly.

The means for C and P are both significantly lower than the means for the other fertilizer treatments. Thus, superphosphate alone has produced no significant difference from control, but the addition of either sulphate of ammonia or sulphate of potash or both to the super has produced a 'significant' increase in green weight.

The adjusted means for the cultural treatments are:

No cultivation, no clover	-	-	-	186.1 lb.
Cultivation only	-	-	-	224.0 lb.
Red clover	-	-	-	188.3 lb.
White clover	-	-	-	183.0 lb.
Red clover + white clover + grass	-	-	-	182.1 lb.

The significant differences at the 5% (1%) levels of probability are 12.6 (16.8), and the standard error is 4.4.

The cultural comparisons show the following:

Treatment 2 (cultivation only) gave a highly significant increase in yield over all other treatments.

The yields from the remaining four treatments (i.e. all those not kept cultivated) did not differ significantly.

TABLE 2. ANALYSIS OF SECOND EXPERIMENTAL CUT
(Using pre-trial weights as concomitant data.)

Source of Variation	d.f.	Adjusted Mean Squares	F	5% (1%) points of F
Squares: rows and columns within squares	35			
Manurial treatments	4	1,590.6	3.43*	2.54(3.68)
Cultural treatments	4	1,928.1	4.15**	2.54(3.68)
Residual	55	464.3		
Total	98			

Coefficient of variation = 10.1%

*Significant at 5%.

**Significant at 1%.

Second Experimental Cut

There were significant differences due to fertilizer treatments. The adjusted treatment means are given below.

C	-	-	-	-	198.5
P	-	-	-	-	214.0
P + N	-	-	-	-	220.0
P + K	-	-	-	-	215.0
P + K + N	-	-	-	-	217.0

The significant differences at the 5% (1%) levels of probability are 13.7 (18.2), and the standard error for all treatments is 4.8.

The fertilizer comparisons gave the following results:

The control yield was significantly lower than that from any fertilizer treatment.

There were no significant differences between the four fertilizer treatments in which superphosphate was used.

For the cultural treatments, the adjusted means are:

No cultivation, no clover	-	-	-	-	209.5
Cultivation only	-	-	-	-	230.5
Red clover	-	-	-	-	212.5
White clover	-	-	-	-	205.0
Red clover, white clover, grass	-	-	-	-	209.0

The SEs and detectable differences are as for fertilizer means.

The cultural comparisons show the following:

The mean for the cultivation treatment was significantly higher than for any other treatment.

There were no significant differences between the remaining treatments.

Total Production for the Two Trial Years

The analysis of co-variance is derived from the green weight (lb.) per plot.

There were highly significant differences due to fertilizer treatments. The adjusted treatment means are given below.

C	-	-	-	380.5
P	-	-	-	398.5
P + N	-	-	-	422.5
P + K	-	-	-	411.5
P + K + N	-	-	-	417.0

The significant differences at 5% (1%) are 20.7 (27.6), and the standard error for all treatments is 7.3.

The fertilizer comparisons are as follows:

The control mean is significantly lower than the phosphate mean at the 8% level of probability, and the phosphate mean in turn is significantly lower than the phosphate and nitrogen mean at the 3% level. There was no significant potash effect.

TABLE 3. ANALYSIS OF BOTH EXPERIMENTAL CUTS
(Using pre-trial weights as concomitant data.)

Source of Variation	d.f.	Adjusted Mean Squares	F	5% (1%) points of F
Squares: rows and columns within squares	35			
Manurial treatments	4	5,701.9	5.35**	2.54 (3.68)
Cultivation treatments	4	14,935.2	14.00**	2.54 (3.68)
Residual	55	1,066.7		
Total	98			

Coefficient of variation = 8.0%

**Significant at 1%.

The means for P + K and P + K + N are both significantly higher than control but neither of them is significantly different from P + N.

There were highly significant differences due to cultivation. The adjusted means are:

No cultivation, no clover	-	-	-	396.0
Cultivation, no clover	-	-	-	454.5
Red clover	-	-	-	401.0
White clover	-	-	-	388.0
Red clover, white clover, grass	-	-	-	391.0

The significant differences at the 5% (1%) levels of probability are 20.7 (27.6), and the standard error for all treatments is 7.3.

The mean for the cultivation treatment was significantly higher than that for any other treatment.

There were no significant differences between the remaining treatments.

DISCUSSION

Fertilizer Treatments

It is clear that fertilizers do have an effect on the growth rate of pampas grass. During the first year of treatment superphosphate plus sulphate of ammonia and sulphate of potash both gave significant increases over the control; no significant response was recorded from the application of superphosphate alone. In the second year of treatment, however, superphosphate induced a significant response and the addition of ammonia and/or potash to it gave no additional response. (The soil on which this trial was conducted was a sandy silt loam close to the Manawatu River and built up from alluvium carried down by it. It is not naturally fertile and there are evidences of potash responses on adjoining pastures.)

Cultural Treatments

Owing to the ingress of volunteer clovers (principally white) into the unsown plots, and a general levelling up of the clover and grass population during the first year of treatment, it has not been possible to compare the effects of clover sowings with a mixture of grasses and clovers and the unsown plots. There is a lack of statistical significance between yields in any of the plots that carried grasses and clovers. The main feature lies in the considerable increase in growth as a result of keeping the ground cultivated and free from ground flora. It has been noted elsewhere* that the effect of grasses and clovers on the yield of pampas is very marked in the early stages of development but becomes progressively smaller as the pampas increases in size. It is possible that, when the pampas begins to lose vitality with age, grass can again become a very important competitive factor.

The efficiency of the analysis of co-variance is worth comment in this trial. Relative to the ordinary analysis of variance, the efficiency values were—1st analysis 288%, 2nd analysis 123%, two-year analysis 233%.

RECOMMENDATIONS

From many previous observations it is clear that clean cultivation has an early beneficial effect on yield. Stands should therefore be planted in cultivated ground and kept clean-cultivated until the first autumn or the spring following planting. It does not seem feasible to continue with cultivation beyond this length of time. Grasses and clovers can be sown between the plants when cultivation ceases in the first autumn or spring after planting and thereafter kept under control by sheep. It is possible (though not taken into account in this trial) that any gain in pampas yield obtained through cultivation over a longer period than this is counterbalanced by the loss of grazing of the ground flora.

The effect of clovers in raising the nitrogen status of the soil and thus increasing pampas yield has not been proved by this trial. In view of the increase in yield of pampas due to the application of nitrogen however, it would seem feasible to suggest that a good clover growth beneath the pampas is of value. Both the clover and the pampas can be stimulated by the application of superphosphate. The addition of sulphate of ammonia would not normally be warranted where a response to superphosphate is obtained, because of the high price of the fertilizer and also the possibility of it suppressing the clovers.

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* Paper by the author in course of preparation.

SCREENING TESTS WITH FUNGICIDES FOR CONTROL OF FRENCH-BEAN RUST (*Uromyces appendiculatus* (Pers.) Unger)

By H. JACKS and R. M. BRIEN, Plant Diseases Division, Department of Scientific and Industrial Research, Auckland.

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Summary

Thirty seven fungicides were tested in the glasshouse for control of *Uromyces appendiculatus* (Pers.) Unger.

Effective compounds were lime sulphur + colloidal sulphur, colloidal sulphur, fine wettable sulphur, zineb, ferbam, maneb, thiram, ziram, captan, dichlone, chloranil, and nitrobenzene.

INTRODUCTION

French-bean rust (*Uromyces appendiculatus* (Pers.) Unger), first observed in New Zealand in 1951, and officially recorded by Brien and Dingley in 1953 has been described previously (Brien and Jacks, 1954). Tests outlined in the present paper were carried out as part of the Division's certification scheme to ascertain the effectiveness of various fungicides in control of this disease.

MATERIALS

Copper compounds: Bordeaux mixture; Cop-ox (50% copper oxychloride) wettable powder; Perenox (50% cuprous oxide) wettable powder; copper zeolite (50% copper) wettable powder; Ialine colloidal copper (15% copper oxychloride) emulsion; Bouisol (15% colloidal copper oxychloride) emulsion; Bouisol + oil (15% colloidal copper oxychloride + 1% white oil) emulsion; and Fungex (8.5% copper ammonium carbonate) liquid.

Sulphur compounds: lime sulphur (15% polysulphides) liquid; Colsul 40 (40% colloidal sulphur) paste; Wetomic sulphur (80% sulphur) wettable powder; and Cosan (75% sulphur) wettable powder.

Dithiocarbamates: Dithane Z 78 (65% zineb) wettable powder; Dithane D 14 (X/Y/Z = xlb. of 25% nabam/ylb. of zinc sulphate / zlb. of hydrated lime) wettable powder; Fernspray (70% ferbam) wettable powder; Manzate (70% maneb) wettable powder; Thirospray (50% thiram) wettable powder; Vancide (30% nabam plus 2 mercapto benzothiazole) wettable powder; Cupram (technical cupric dimethyl dithiocarbamate) powder; Zirospray (70% ziram) wettable powder; and Fuciasin ultra (70% ziram) wettable powder.

Miscellaneous compounds: Flit 406 (50% captan) wettable powder; Phygon XL (50% dichlone) wettable powder; Spergon w.p. (48% chloranil) wettable powder; Shirlan A.G. (25% salicylanilide) paste; Crag 341C (34% glyodin) liquid; Crag 5379 (75% 1, 2, 3 trithio-5, 8 diazocyclonone-4, 9 dithione) liquid; Crag 5400 (75% alpha, alpha trithio bis (n-dimethyl thiophosphoramidate) liquid; Crag 658 (15% cuprous oxide + 10% zinc oxide + 6% chromium oxide) liquid; Crag 1182 (3.8% 4-chloro-3, 5-dimethylenoxy ethanyl) wettable powder; Nirit (15% dinitrobenzene) wettable powder; 8 quinoline, powder; 8

quinolinol sulphate, liquid; 8 quinolinol benzoate, powder; zinc 8 quinolinolate, powder; copper 8 quinolinolate, powder; and 36 L (98% 1 - (p - sulphanylephenyl) - 3 : 5 - dimethyl - 4 - nitrosopyrazole) powder.

EXPERIMENTAL METHODS

Seed of French bean, variety Fardenlosa, was sown in 4-in. pots, one seed per pot. When the two primary leaves had fully developed, the growing tips were removed and four plants for each treatment replication were sprayed with 80 ml. of the fungicidal suspensions. Twenty-four hours later, plants were inoculated by spraying with a rust-spore suspension containing 25,000 to 30,000 spores per ml. and placed in a humidity cabinet (92% $\pm$ 1% relative humidity at 63°F. $\pm$ 1°F.) for 24 hours). Plants were then moved to a glasshouse held at approximately 70°F. Lesion counts were made 10 to 14 days after inoculation and figures in the tables give the means from two leaves on each of the four test plants used per treatment.

Any material used in the first trial (Table 1) that was not significantly different in efficacy from those materials giving complete control was retested (Table 2). Materials that gave the best results in these two tests were used in the final selection test (Table 2).

RESULTS

Under these test conditions satisfactory control was obtained with:—

Lime sulphur at 0.5% combined with colloidal sulphur at 0.08% (Colsul 40 at 0.2%); colloidal sulphur at 0.4% (Colsul 40 at 1%); and fine wettable sulphur at 0.38% (Cosan at 0.5%).

Zineb in a tank mix formulation of nabam at 0.19% (Dithane D14 at 0.1% + zinc sulphate + hydrated lime); ferbam at 0.07% (Fermispray at 0.1%); maneb at 0.018% (Manzate at 0.025%); thiram at 0.2% (Thirospray at 0.4%); and ziram at 0.28% (Zirospray and Fuclasin Ultra at 0.4%).

Captan at 0.2% (Flit 406 at 0.4%); dichlone at 0.013% (Phygon XL at 0.025%); chloranil at 0.012% (Spergon w.p. at 0.025%); and nitrobenzene at 1% (Nirit at 0.67%).

Nabam + mercapto benzothiazole (Vancide) gave satisfactory control at all concentrations tested but caused plant damage.

Zineb at 0.13% used in a proprietary formulation (Dithane Z78 at 0.2%) was effective in 4 out of 6 tests.

As in tests carried out with broad-bean rust (*Uromyces fabae* (Pers.) de Bary) (Jacks, 1954) copper compounds allowed a relatively large number of rust lesions to develop. Also, the coarser wettable sulphur (Wetomic) was not as effective as the finer sulphur (Cosan).

In tests here reported some differences were observed from results obtained with *U. fabae* as test organism. Thus zineb in tank formulation was more effective than the proprietary compound, the two formulations of ziram under test were equally effective, and lime sulphur alone did not satisfactorily control rust.

TABLE 1. EFFECT OF FUNGICIDES APPLIED AT SEVERAL CONCENTRATIONS ON

Copper Compounds				Sulphur Compounds			
Treatment	Amts. in 100 gal. water	Lesions per leaf		Treatment	Amts. in 100 gal. water	Lesions per leaf	
		Mean No.	log (x + 1)			Mean No.	log (x + 1)
Check	—	720.3	2.86	Check	—	588.5	2.76
Bordeaux mixture	10 lb: 8 lb.	85.5	1.92	Lime sulphur*	2 gal.	0.0	0.00
"	6 lb: 8 lb.	73.4	1.86	" "	1 gal.	0.6	0.17
Cop-ox	10 lb.	84.7	1.93	" "	0.75 gal.	0.8	0.23
"	5 lb.	48.0	1.70	" "	0.5 gal.	5.5	0.81
"	2.5 lb.	38.0	1.58	" "	0.25 gal.	19.1	1.30
Perenox	10 lb.	19.9	1.31	Lime sulphur + Colsul 40*	1 gal: 2 lb.	0.0	0.00
"	5 lb.	15.3	1.22	" "	1 gal: 1 lb.	0.0	0.00
"	2.5 lb.	17.5	1.27	" "	0.75 gal: 2 lb.	0.0	0.00
Copper zeolite	10 lb.	749.9	2.87	" "	0.75 gal: 1 lb.	0.1	0.03
" "	5 lb.	886.5	2.95	" "	0.5 gal: 2 lb.	0.2	0.05
" "	2.5 lb.	770.6	2.89	" "	0.25 gal: 2 lb.	0.1	0.04
Ialine Col. Copper*	4 gal.	403.1	2.65	Colsul 40	16 lb.	0.0	0.00
"	2 gal.	226.3	2.35	"	12 lb.	0.0	0.00
"	1 gal.	315.0	2.50	"	10 lb.	0.4	0.13
"	0.5 gal.	320.1	2.50	"	8 lb.	3.9	0.68
Bouisol*	4 gal.	9.1	1.00	"	4 lb.	2.3	0.51
"	2 gal.	10.9	1.07	"	2 lb.	7.8	0.94
"	1 gal.	33.1	1.53	Wetomic sulphur	16 lb.	11.4	1.10
"	0.5 gal.	36.3	1.58	"	12 lb.	12.3	1.11
Bouisol + oil*	4 gal.	369.1	2.57	"	10 lb.	14.1	1.18
" *	2 gal.	630.5	2.80	"	8 lb.	12.8	1.14
"	1 gal.	140.4	2.16	"	4 lb.	20.2	1.32
Fungex	0.5 gal.	17.3	1.27	"	2 lb.	75.5	1.89
"	0.25 gal.	102.5	2.02	Cosan	16 lb.	0.0	0.00
				"	12 lb.	0.0	0.00
				"	10 lb.	0.0	0.00
				"	8 lb.	0.0	0.00
				"	6 lb.	0.2	0.05
				"	5 lb.	0.1	0.03
				"	4 lb.	4.5	0.75
				"	2 lb.	6.4	0.84

DEVELOPMENT OF FRENCH-BEAN RUST (Means from two replications, each of four plants)

Thiocarbamates				Miscellaneous Compounds			
Treatment	Amts. in 100 gal. water	Lesions per leaf		Treatment	Amts. in 100 gal. water	Lesions per leaf	
		Mean No.	log (x + 1)			Mean No.	log (x + 1)
Check	—	987.6	2.98	Check	—	782.6	2.89
Dithane Z78*	6 lb.	0.0	0.00	Flit-406	6 lb.	0.5	0.14
"	4 lb.	0.0	0.00	"	4 lb.	0.3	0.06
"	2 lb.	2.0	0.47	"	2 lb.	3.9	0.70
"	1 lb.	3.6	0.65	"	1 lb.	11.1	1.07
Dithane D14*	6 lb: 3 lb. 1 lb.	0.0	0.00	Phygon XL*	1 lb.	0.0	0.00
"	4 lb: 2 lb. 1 lb.	0.0	0.00	"	0.5 lb.	0.0	0.00
"	2 lb: 1 lb. 0.5 lb.	0.1	0.02	"	0.25 lb.	0.5	0.12
"	1 lb: 0.5 lb. 0.25 lb.	0.3	0.10	Spergon w.p.	1 lb.	0.0	0.00
Fernspray	6 lb.	0.0	0.00	"	0.5 lb.	0.0	0.00
"	4 lb.	0.0	0.00	"	0.25 lb.	0.4	0.12
"	2 lb.	0.3	0.08	Shirlan A.G.*	6 lb.	2.4	0.53
"	1 lb.	0.7	0.20	"	4.5 lb.	42.5	1.63
Manzate*	6 lb.	0.0	0.00	Crag 341C	2 lb.	10.7	1.07
" *	4 lb.	0.0	0.00	"	1 lb.	24.7	1.41
" *	2 lb.	0.0	0.00	"	0.5 lb.	38.5	1.60
"	1 lb.	0.0	0.00	Crag 5379	2 lb.	32.9	1.53
Thiросpray	8 lb.	0.1	0.03	"	1 lb.	156.4	2.20
"	4 lb.	0.3	0.08	"	0.5 lb.	418.2	2.62
"	2 lb.	1.4	0.37	Crag 5400	2 lb.	2.8	0.56
"	1 lb.	1.3	0.35	"	1 lb.	4.9	0.75
Vancide*	14 lb.	0.0	0.00	"	0.5 lb.	149.8	2.18
" *	7 lb.	0.0	0.00	"	2 lb.	461.2	2.67
" *	3.5 lb.	6.1	0.86	"	1 lb.	834.9	2.92
" *	1.8 lb.	3.5	0.64	"	0.5 lb.	1000+	3.00
Cupram	4 lb.	20.4	1.33	Crag 1182	2 lb.	875.0	2.94
"	2 lb.	12.8	1.13	"	1 lb.	950.0	2.98
"	1 lb.	19.5	1.31	"	0.5 lb.	1000+	3.00
"	0.5 lb.	28.3	1.47	Nirit	13.4 lb.	0.0	0.00
Zirospray	6 lb.	0.0	0.00	"	6.7 lb.	0.1	0.02
"	4 lb.	0.4	0.11	"	3.4 lb.	23.0	1.38
"	2 lb.	1.4	0.38	8 Quinolinol*	1 lb.	82.5	1.93
"	1 lb.	11.4	1.09	8 Quinolinol sulphate*	1 lb.	255.8	2.41
Fuclasin Ultra	6 lb.	0.0	0.00	8 Quinolinol benzoate*	1 lb.	35.5	1.56
"	4 lb.	0.6	0.17	Zinc 8 quino- linolate	1 lb.	21.8	1.35
"	2 lb.	0.8	0.24	Copper 8 quinolinolate*	1 lb.	11.7	1.10
"	1 lb.	1.0	0.30	36L	1 lb.	40.0	1.60

*Fungicides at this strength caused necrotic spotting on leaves.

TABLE 2. RESULTS FROM THE MOST EFFECTIVE TREATMENTS IN PREVENTION OF FRENCH-BEAN RUST

(Means from two replications each of four plants)

Treatments selected from Table 1				Final Selection			
Treatment	Amts. in 100 gal. water	Lesions per leaf		Treatment	Amts. in 100 gal. water	Lesions per leaf	
		Mean No.	Log (x + 1)			Mean No.	Log (x + 1)
Check	—	710.6	2.85	Check	—	819.6	2.92
Lime sulphur	1 gal	1.2	0.35	Lime sulphur:	0.75 gal:		
" "	0.75 gal.	5.0	0.79	Colsul 40	2 lb.	0.0	0.00
Lime sulphur	0.75 gal. +			" "	0.5 gal: 2 lb.	0.0	0.00
+ Colsul 40	2 lb.	0.5	0.13	Cosan	10 lb.	0.0	0.00
" "	0.5 gal. +			Colsul 40	5 lb.	0.1	0.03
" "	2 lb.	0.7	0.20	Dithane Z78	2 lb	0.5	0.14
" "	0.25 gal. +			Dithane D14	1 lb: 0.5 lb:		
" "	2 lb	2.1	0.50		0.25 lb.	0.0	0.00
Colsul 40	12 lb.	0.0	0.00	Fernspray	1 lb.	0.4	0.11
"	10 lb.	0.5	0.14	Manzate	0.5 lb.	0.1	0.02
Cosan	6 lb.	0.1	0.03	Thirospray	4 lb.	0.0	0.00
"	5 lb.	0.3	0.10	Zirospray	4 lb.	0.5	0.15
Dithane Z78	4 lb.	0.0	0.00	Fuclasin Ultra	4 lb.	0.3	0.11
"	2 lb.	0.3	0.08	Flit-406	4 lb.	0.0	0.00
Dithane D14*	2 lb: 1 lb:			Phygon XL	0.25 lb.	0.1	0.03
"	0.5 lb.	0.0	0.00	Spergon w.p.	0.25 lb.	0.6	0.15
"	1 lb: 0.5 lb.			Nirit	6.7 lb.	0.0	0.00
"	0.25 lb.	0.3	0.09				
Fernspray	1 lb.	0.3	0.09				
"	0.5 lb.	0.8	0.26				
Manzate	0.5 lb.	0.0	0.00				
"	0.25 lb.	0.2	0.07				
Thirospray	4 lb.	0.0	0.00				
"	2 lb.	1.0	0.31				
Zirospray	4 lb.	0.0	0.00				
"	2 lb.	2.4	0.52				
Fuclasin Ultra	4 lb.	0.0	0.00				
" "	2 lb.	1.7	0.46				
Flit 406	4 lb.	0.2	0.05				
"	2 lb.	5.0	0.79				
Phygon XL	0.5 lb.	0.0	0.00				
"	0.25 lb.	0.3	0.08				
Spergon w.p.	0.5 lb.	0.1	0.03				
"	0.25 lb.	0.3	0.10				
Nirit	6.7 lb.	0.1	0.02				
"	5.0 lb.	2.8	0.57				

Difference required for significance at 1%: 0.25

*Fungicide at this strength caused necrotic spotting on leaves.

The following materials, at concentrations tested, did not give satisfactory control of the rust: cupram; salicylanilide (Shirlan A.G.); glyodin (Crag 341C); Crag 5379; Crag 5400; Crag 658; Crag 1182; 8 quinolinol; 8 quinolinol sulphate; 8 quinolinol benzoate; zinc 8 quinolinolate; copper 8 quinolinolate; and 36 L.

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ADJUSTMENT OF CASEIN/FAT RATIO OF MILK FOR CHEESEMAKING

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Summary

A rapid method for standardizing milk for cheesemaking is described. The method reduces the delay that occurs when testing of milk and adjustment of fat content take place after the cheese vat has been filled and before cheesemaking can commence.

Samples for determination of fat and casein (by Gerber and Walker methods) are taken from a receiving vat which is large enough to give reasonable bulking of milk supplies as received. Each sample represents sufficient milk to fill one or two cheese vats. During the filling of each vat, part of the milk is diverted through a cream separator, the skim milk being returned to the line carrying whole milk to the cheese vat. The rate of delivery (lb. per min.) of butterfat as cream from the separator having been previously determined, the time during which milk should be bypassed through the separator while filling a vat to a calibration mark can be arrived at.

A formula is deduced, and from it tables are made up which give the weight of butterfat to be removed while filling a vat of known capacity to bring the milk to a given casein/fat ratio.

The relationship between casein/fat ratio in milk and fat in the water-free solids (F.W.F.S.) in cheese is discussed. The F.W.F.S. of cheese from milk of a given casein/fat ratio has a standard deviation of the order of 0.6. The variation is attributed to changes in the ratio of Walker casein to true casein in milk, to changes in whey losses and in cheese composition, and to sampling and analytical errors in determination of F.W.F.S. in cheese.

Only small additional variations are introduced by the rapid method of standardization, provided that the milk received is sufficiently bulked before sampling. Even under the best conditions, however, if the number of cheeses with F.W.F.S. falling below the minimum is to be kept small, the average F.W.F.S. in the standardized cheese must be at least 1% above the minimum allowed.

INTRODUCTION

In the manufacture of cheese that is to be standardized to a given content of fat in the water-free solids (F.W.F.S.) it is necessary to add a calculated quantity of skim milk to whole milk so as to bring the casein/fat ratio to the value judged likely to give the desired F.W.F.S. in the cheese. In the method generally adopted (e.g. McDowall and Valentine¹) the cheese vat is partly filled with whole milk, which is then tested for its content of fat and casein (or of fat only). From these figures the volume of skim milk to be added is calculated. The skim milk is obtained by separation of a further quantity of whole milk. Since all these operations have to be completed before cheesemaking can be begun, a considerable delay results.

A method that considerably reduces the time taken in standardization was developed by one of us (W.L.H.) during the course of a commercial-scale trial of the manufacture of "fat-adjusted" cheese (F.W.F.S. not less than 52% carried out in collaboration by the Dairy Division of the

Department of Agriculture and the Dairy Research Institute. In this method the milk is sampled in the receiving vat while supplies are arriving. Fat is removed by separation from some of the milk while the cheese vat is being filled. Some loss of accuracy in the standardization may be involved, but the error is small in comparison with the other random variations that affect the F.W.F.S. of the cheese.

PRACTICAL DETAILS OF METHOD OF STANDARDIZATION

Milk delivered by individual suppliers during the two-hour receiving period is run, after weighing, into a large (say 2,000-gal.) receiving vat. It is pumped continuously from this vat through the pasteurizer to the cheese vats, each of which is filled in turn to a calibration mark. The receiving vat is kept as full as possible in order to provide the maximum degree of bulking of the individual milk supplies. At suitable intervals, the milk in the receiving vat is thoroughly stirred and a sample is taken and tested for fat by the Gerber method and for casein by the Walker method. For the purposes of standardization it is assumed that the sample is representative of the milk delivered to one or two cheese vats, i.e. any change in composition due to the addition of further supplies to the receiving vat during the filling of these vats is neglected.

While each vat is being filled, some of the milk is diverted through a cream separator for a sufficient time to remove a calculated quantity of fat. The skim milk is returned to the milk line for delivery to the cheese vat along with the whole milk. The amount of fat to be removed is computed from the fat and casein tests on the milk sample and the volume of milk to be contained in the vat when filled to the mark. The formula to be used is discussed later.

The quantity of fat removed as cream may be measured by weighing the cream obtained from the separator, the fat content of the cream being assumed to be the same as that found by actual testing in an earlier run under similar conditions. A more accurate and convenient method, however, is based on the fact that a separator running at full capacity on milk of uniform fat content will deliver as cream a constant weight of fat per minute independent of the fat test of the cream. The quantity of fat removed may thus be controlled simply by timing the period during which milk is bypassed through the separator. The rate of delivery of butterfat by the separator must be determined with the skim milk and cream outlet settings to be used and with milk of known fat content. If the samples from the receiving vat show an appreciably different fat test from that of the milk on which the separator was calibrated, a proportional adjustment must be made in the time during which milk is run through the separator. The calibration should be repeated at intervals.

The accuracy of this method was confirmed in practice. It was found that with a 660-gallon-per-hour separator in a series of runs giving cream varying from 20% to 49% in fat content, the rate of delivery of butterfat varied only from 4.9 to 5.2 lb. per minute. The day-to-day variation when running on bulk milk in one factory was negligible.

CALCULATION OF QUANTITY OF BUTTERFAT TO BE REMOVED TO BRING MILK TO A GIVEN CASEIN/FAT RATIO

A number of formulae for use in standardization of milk to a given casein/fat ratio have been discussed by Cox². In general, they have been intended for use where only the fat is to be determined on the milk received. The casein content is assumed to have some definite relationship to the fat content. Cox (*loc. cit.*) has derived a formula for use where the fat and solids-not-fat in the milk have been determined and where the composition of the cheese to be made and the losses of fat and solids-not-fat to be expected in manufacture are known.

As there may be an appreciable variation in the casein content of milk of a given fat content, milk can be standardized much more accurately where both casein and fat contents are known than where only fat has been estimated. A method using the Walker Casein Test has been advocated by McDowall and Valentine¹, Gilmore and Price³, and others. This method was followed in the present investigation.

Where milk is to be standardized by addition of a measured quantity of skim milk, the calculation is a matter of simple proportion if the slight change in casein content due to removal of some of the fat from the milk is neglected. McDowall and Valentine¹ published formulae and tables for use in standardizing milk by this method. Where the quantity of cream or butterfat removed is to be measured as in the method now recommended, allowance must be made for the casein contained in the cream serum. A formula may be developed as follows:

Let

$$\text{Wt. of standardized milk required (lb.)} = W$$

$$\text{Fat in original whole milk (\%)} = f$$

$$\text{Casein in original whole milk (\%)} = c$$

$$\text{Fat in cream (\%)} = F$$

$$\text{Casein/fat ratio desired} = R$$

$$\text{Wt. of butterfat to be removed (lb.)} = x$$

Then

$$\text{Wt. of cream removed (lb.)} = \frac{100x}{F}$$

$$\text{Wt. of serum in cream (lb.)} = x \left(\frac{100-F}{F} \right)$$

$$\text{Casein in serum of milk or cream (\%)} = \frac{100c}{100-f}$$

$$\text{Wt. of casein in cream serum (lb.)} = \frac{x}{100} \left(\frac{100-F}{F} \right) \frac{100c}{100-f}$$

$$\therefore \frac{cx}{F} \left(\frac{100-F}{100-f} \right)$$

$$\text{Wt. of whole milk (lb.)} = W + \frac{100x}{F}$$

$$\begin{aligned} \text{Wt. of fat in standardized milk (lb.)} &= \frac{f}{100} \left(W + \frac{100x}{F} \right) - x \\ &= \frac{fW}{100} + \frac{fx}{F} - x \end{aligned}$$

$$\begin{aligned} \text{Wt. of casein in standardized milk (lb.)} &= \frac{c}{100} \left(W + \frac{100x}{F} \right) - \frac{cx}{F} \left(\frac{100-F}{100-f} \right) \\ &= \frac{cW}{100} + \frac{cx}{F} \left(1 - \frac{100-F}{100-f} \right) \end{aligned}$$

Casein/fat in standardized milk

$$\begin{aligned} &= \frac{\frac{cW}{100} + \frac{cx}{F} \left(1 - \frac{100-F}{100-f} \right)}{\frac{fW}{100} + \frac{fx}{F} - x} = R \\ x &= \frac{\frac{W}{100} (fR - c)}{R - \frac{fR}{F} + \frac{c}{F} \left(1 - \frac{100-F}{100-f} \right)} \\ &= \frac{\frac{WF}{100} \left(f - \frac{c}{R} \right)}{F - f + \frac{c}{R} \left(1 - \frac{100-F}{100-f} \right)} \end{aligned}$$

Where $W = 10,000$ lb. and $F = 40\%$

$$x = \frac{4,000 \left(f - \frac{c}{R} \right)}{40 - f + \frac{c}{R} \left(1 - \frac{60}{100-f} \right)}$$

Tables 1, 2, and 3 have been calculated for these values of W and F for casein/fat ratios of 0.65, 0.68, and 0.70. The tables can be used with only a small error for cream of fat content from 35% to 45% since the slight increase or decrease in the amount of casein removed in the cream serum can have little effect on the composition of the standardized milk.

e.g. in milk to be standardized:

$$\begin{array}{lll} f = 4.5 & c = 2.5 & R = 0.68 \\ \text{For } F = 35\% & x = 90.9 \text{ lb.} & \\ F = 40\% & x = 89.6 \text{ lb.} & \\ F = 45\% & x = 88.1 \text{ lb.} & \end{array}$$

For all these cases the value of 90 given for x in the tables could be taken without causing a significant error in the fat content of the cheese.

FAT CONTENT OF STANDARDIZED MILK

If the change in casein concentration resulting from removal of some of the fat from the milk is neglected,

$$\text{Fat in standardized milk (\%)} = f_s = c/R.$$

More accurately,

$$\begin{aligned} f_s &= \left[\frac{f}{100} \left(W + \frac{100x}{F} \right) - x \right] \frac{100}{W} \\ &= f + \frac{100fx}{FW} - \frac{100x}{W} \\ &= f - \frac{100x}{FW} (F - f) \end{aligned}$$

Where $F = 40$ and $W = 10,000$ lb.

$$f_s = f - \frac{x}{4,000} (40 - f).$$

In view of the considerable variations which, as shown later, affect the F.W.F.S. of cheese, it may seem that an approximate standardization formula would suffice. Such a formula would be likely, however, to give a systematic error that would affect the F.W.F.S. of all the cheese in the same direction. The more accurate formula is therefore to be preferred and, if tables are used, involves no more work.

CHOICE OF CASEIN/FAT RATIO TO WHICH MILK SHOULD BE STANDARDIZED

It is not possible to specify exactly the casein/fat ratio for milk that will yield cheese with a given F.W.F.S. The F.W.F.S. in cheese made from milk of a certain Walker-casein/fat ratio will depend on:

1. The ratio of Walker casein to true cheese protein in the milk.
2. The losses of fat and solids-not-fat in the whey.
3. The proportion of salt and solids other than fat and protein in the cheese.

The error in determination of the F.W.F.S. in cheese must also be taken into account.

McDowall⁴ published data for the casein/fat ratio in milk and the F.W.F.S. in cheese for 785 vats in a number of different factories. These results have been shown in graphical form in Fig. 1. In order to obtain a more nearly linear relationship than could result from plotting F.W.F.S. against casein/fat ratio, the F.W.F.S. has been plotted against fat/(casein + fat). (The casein/fat ratios have also been indicated on a separate scale.) This method would be expected to yield an approximately linear graph which would pass through the origin. The mean value of F.W.F.S. for each group of vats with the same fat/(casein + fat) ratio has been shown as a point. Maximum and minimum values found for each group are connected by light zig-zag lines. The heavy broken line passes through the origin and through the mean for all the

Fat	Casein												
	2.0	2.1	2.2	2.3	2.4	2.5	2.6	2.7	2.8	2.9	3.0	3.1	3.2
3.2	13												
3.3	24	7											
3.4	34	18	2										
3.5	45	28	12										
3.6	56	39	23	6									
3.7	66	50	34	17	1								
3.8	78	61	44	28	12								
3.9	88	72	55	39	22	6							
4.0	99	83	66	50	33	16							
4.1	110	94	77	60	44	27	11						
4.2	122	105	88	71	54	38	22	5					
4.3	133	116	99	82	66	49	32	16					
4.4	144	127	110	93	76	60	43	27	10				
4.5	155	138	121	104	88	71	54	38	21	4			
4.6	167	150	132	116	99	82	65	48	32	15			
4.7	178	161	144	127	110	93	76	59	43	26	9		
4.8		172	155	138	121	104	87	70	54	37	20	3	
4.9			167	150	132	116	98	82	65	48	31	14	
5.0				161	144	127	110	93	76	59	42	25	8
5.1				172	155	138	121	104	87	70	53	36	19
5.2					167	149	132	115	98	81	64	47	30
5.3					178	161	144	127	110	92	75	58	41
5.4						172	155	138	121	104	86	70	52
5.5							167	150	132	115	98	81	64
5.6							179	161	144	126	109	92	75
5.7								173	155	138	121	103	86
5.8								184	167	150	132	115	97
5.9									179	161	144	126	109
6.0										173	155	138	120

2.0 2.1 2.2 2.3 2.4 2.5 2.6 2.7 2.8 2.9 3.0 3.1 3.2

Casein

Fat	Casein												
	2.0	2.1	2.2	2.3	2.4	2.5	2.6	2.7	2.8	2.9	3.0	3.1	3.2
3.2	36	21	6										
3.3	47	32	17	2									
3.4	58	42	27	12									
3.5	68	53	38	23	8								
3.6	79	64	49	33	18	3							
3.7	90	75	59	44	29	14							
3.8	101	86	70	55	40	25	9						
3.9	112	97	81	66	51	35	20	5					
4.0	123	108	92	77	61	46	31	15					
4.1	135	119	103	88	72	57	41	26	11				
4.2	146	130	114	99	83	68	52	37	21	5			
4.3	157	141	126	110	94	79	63	48	32	17			
4.4	168	153	137	121	105	90	74	59	43	28	12		
4.5	180	164	148	132	117	101	85	70	54	39	23	8	
4.6	191	175	159	144	128	112	96	81	65	50	34	19	3
4.7	203	187	171	155	139	123	108	92	76	61	45	30	14
4.8		198	182	167	150	134	119	103	87	72	56	40	25
4.9			194	178	162	146	130	114	99	83	67	51	36
5.0				189	173	157	141	126	110	94	78	62	47
5.1				201	185	169	153	137	121	105	89	74	58
5.2					197	181	164	148	132	116	101	85	69
5.3					208	192	176	160	144	128	112	96	80
5.4						204	188	171	155	139	123	107	91
5.5							199	183	167	151	135	119	103
5.6							211	195	178	162	146	130	114
5.7								207	190	174	158	142	125
5.8									218	202	186	170	153
5.9										214	198	181	165
6.0											209	193	177

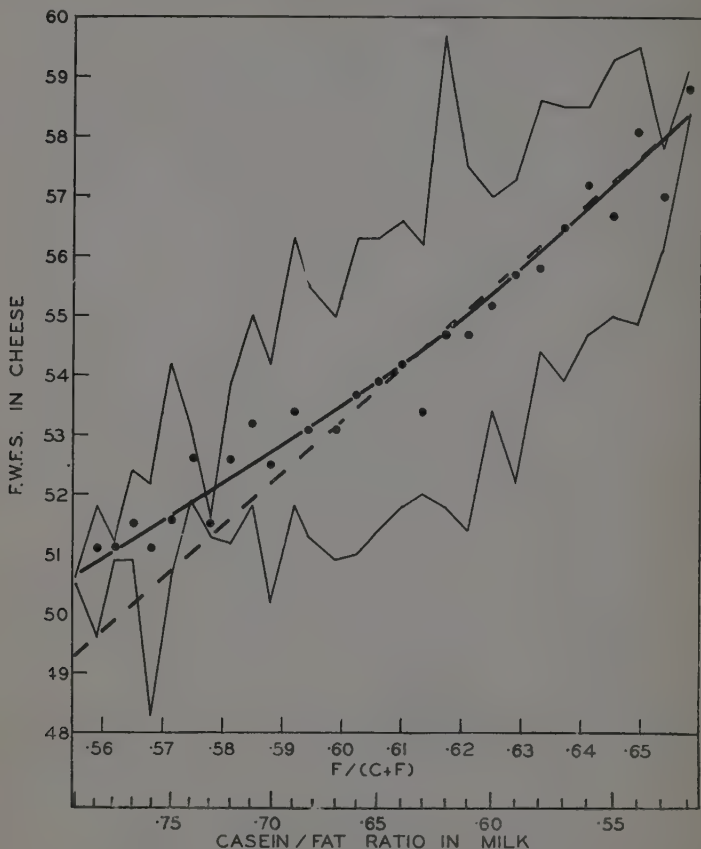


Fig. 1.—Relation between F.W.F.S. in cheese and casein/fat ratio in milk (after McDowall).

vats. The points for values of $F/(C+F)$ above 0.60 lie fairly close to the line but those for lower values tend to diverge, as indicated by the continuous heavy line. As the number of vats with low $F/(C+F)$ ratios was small and the spread of F.W.F.S. values was considerable, this divergence is not necessarily significant.

It has been found in practice that the casein/fat ratio to which milk should be standardized to obtain cheese of a given F.W.F.S. varies in different districts. In one factory it may vary during the season. Data from the two factories in which the present experiments were made confirm this observation. In Fig. 2 have been plotted monthly averages for F.W.F.S. in cheese against $F/(C+F)$ and C/F for the milk. Estimations of casein and fat in the standardized milk were made by the factory staff. F.W.F.S. in the cheese was determined at the Grading

Stores. In each factory during any one month the casein/fat ratio of the standardized milk was constant within the experimental error of standardization, but the ratio was altered at intervals during the season whenever the F.W.F.S. results indicated that such a change was necessary.

The standard curve of Fig. 1 has been drawn in Fig. 2 in order to allow comparison of results at different casein/fat ratios. For Factory A the average F.W.F.S. for the first three months of the season was well above the line. Points for later months cluster fairly closely round the line. The indications are that in this factory the milk should have been standardized to a higher casein/fat ratio in the spring than later in the season.

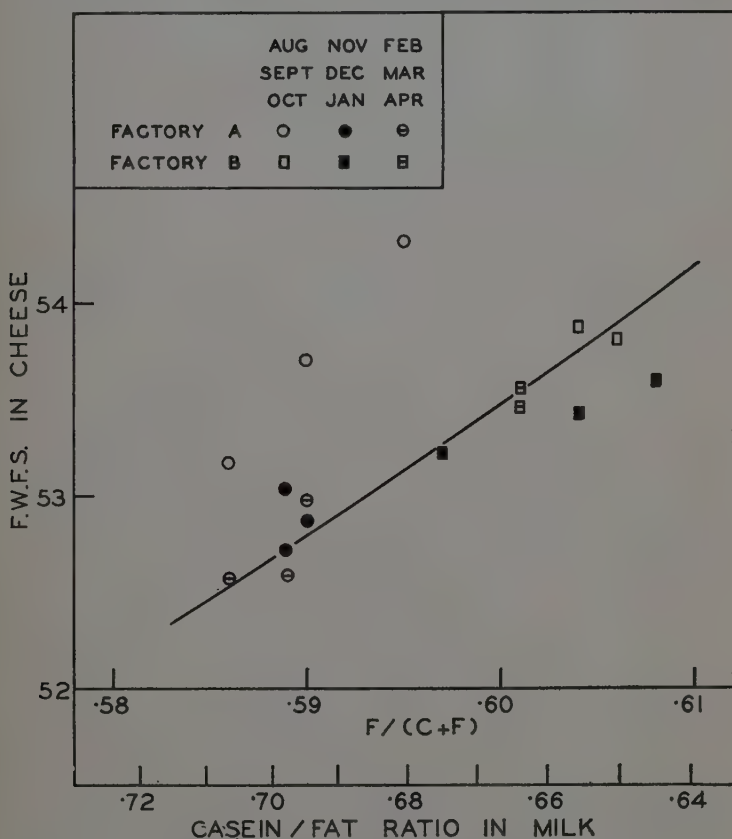


Fig. 2.—Relation between F.W.F.S. in cheese and casein/fat ratio in milk for Factories A and B.

The average values for Factory B show less deviation from the line, the greatest divergence being in the months of November and December when the points were appreciably below the line. During April and May the cheese from this factory showed values of F.W.F.S. which

were very low for the casein/fat ratio concerned. Since only a small number of vats were involved, however, these values were not considered a reliable average and have not been included on the graph.

VARIATION IN F.W.F.S. OF CHEESE FROM MILK OF A GIVEN CASEIN/FAT RATIO

Where analyses of single cheeses instead of averages for large numbers of cheeses are considered, the error of sampling and analysis in determination of F.W.F.S. becomes important. McDowall⁵ found variations of 1.5 in the F.W.F.S. figure in samples from different parts of a single cheese (std. dev. 0.4) and of 1.0 (std. dev. 0.2) in single samples from different cheeses from one vat. He estimated the possible error of actual determination at 1.5 and concluded that the F.W.F.S. estimation could give a maximum error of 4.5 though such divergence would be seldom met.

An example of the variations found in practice has been given by McDowall¹ who tabulated the F.W.F.S. values found for cheese from 202 vats of milk all with a casein/fat ratio of 0.70. These data are summarized in Table 5 together with results from the two factories in the present investigation.

As the data for Factories A and B cover a whole season, they will include the seasonal variations noted earlier. This will, however, be responsible for only a small part of the total variation.

TABLE 4. F.W.F.S. IN CHEESE FROM MILK OF A GIVEN CASEIN/FAT RATIO

Source of Data	No. of Vats	Casein/Fat Ratio in Milk	F.W.F.S. in Cheese	
			Mean	Std. Dev.
McDowall ⁴	202	0.70	53.32	0.63
Factory A	218	0.69	53.25	0.65
	290	0.70	53.00	0.45
	207	0.71	52.93	0.40
	78	0.66	53.42	0.65
Factory B	48	0.67	53.35	0.70
	61	0.68	53.27	0.79

If 0.6 is taken as a typical standard deviation of the F.W.F.S. of cheese from milk of a given casein/fat ratio, one-third of the cheese could be expected to come either above or below the limits of 0.6 from the average F.W.F.S., the numbers above and below being approximately equal, i.e. the F.W.F.S. of one-sixth of the cheese would fall below the average for all the cheeses by more than 0.6. Approximately 95% of the cheeses would have an F.W.F.S. within 1.2 of the mean and only 2.5% would fall below this range. Thus if the minimum F.W.F.S. permissible for the cheese is 52% and no errors in standardization

occurred, some 21½% of the cheese would have an F.W.F.S. below the minimum of 52% where a safety margin of 1.2% was allowed and the average F.W.F.S. of all the cheese was 53.2%.

RANGE OF RESULTS FOUND IN PRACTICE

A convenient means of studying the accuracy of control of F.W.F.S. in the cheese and the safety margin that should be allowed is that of Statistical Quality Control^{6, 7, 8}. To use this method it is necessary to draw "samples", each usually containing 3, 4, or 5 items, at regular intervals during production. From the variation between and within samples "control limits" can be calculated and any variations in conditions affecting the quality of the product can be detected.

For the first example of the application of this method a period has been taken extending over two months, during which Factory A was making 16 vats of cheese a day. Each "sample" consists of cheese from four successive vats. The "control chart" is shown in Fig. 3. The

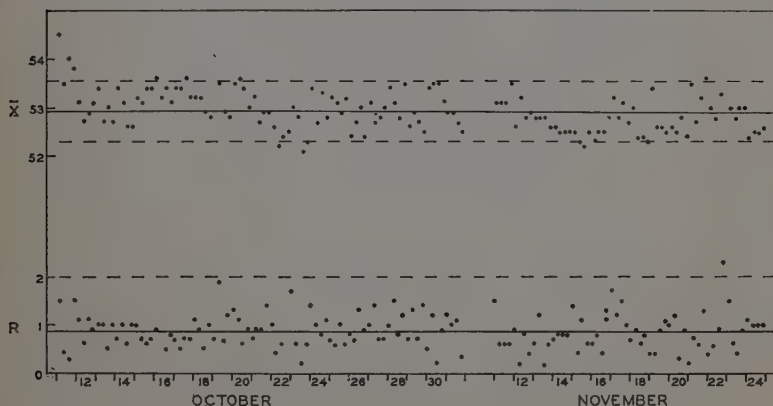


Fig. 3.—Control chart for Factory A.

$$M_x = 52.93 \quad \bar{R} = 0.87$$

$$\text{Limits for } \bar{X} \text{ (average of 4 vats)} = 52.93 \pm 0.635$$

$$\text{Limits for individual vats} = 52.93 \pm 1.27$$

$$\text{Limits for } R = 0, \quad 1.99$$

mean F.W.F.S. for each sample of 4 vats is plotted in the upper part of the chart as $\bar{X}$. The range of F.W.F.S. values in each sample is plotted below as R . The continuous horizontal lines represent M_x (mean value of X for the whole period) and $\bar{R}$ (mean value of R) respectively. The broken lines indicate the control limits, which are calculated as follows:—

The range ($\bar{R}$) is used to estimate the standard deviation (σ). (Values of the ratio $\bar{R}/\sigma$ for samples each of a given number of observations are available in statistical tables.) The control limits are

taken as $\pm 3\sigma$ and should include 99.75% of the samples. Each of the limits can thus be calculated by multiplying $\bar{R}$ by an appropriate constant:

$$\text{Limits for } \bar{X} \text{ chart} = \bar{X} \pm A_2\bar{R}$$

$$\text{Limits for individual observations} = \bar{X} \pm I_2\bar{R}$$

$$\text{Lower limit for R chart} = D_3\bar{R}$$

$$\text{Upper limit for R chart} = D_4\bar{R}$$

Values of these constants for a few values of n are:

n	A_2	I_2	D_3	D_4
3	1.023	1.77	0	2.574
4	0.720	1.46	0	2.282
5	0.577	1.20	0	2.114

The control chart for Factory A (Fig. 3) illustrates that constant conditions operated throughout the period since very few points are "out of control" and there is no trend either upwards or downwards.

The variation of control limits from the mean was 0.635 for samples of 4 vats or 1.27 for individual vats. This corresponds to a standard deviation of only 0.42 in F.W.F.S. which is less than most of the standard deviations given in Table 4 for F.W.F.S. of cheese from milk of given casein/fat ratio. The errors of standardization must therefore have been small in comparison with the other variations.

In order to ensure that no vats (except those that were out of control) gave cheese with F.W.F.S. below 52% it would have been necessary to allow a safety margin of 1.27%. The actual margin allowed was 0.93% and only 5 vats out of 656 gave a F.W.F.S. below 52%.

The accurate standardization at Factory A was made possible by the fact that the individual milk supplies differed little in fat content and that the receiving vat was large (2,500 gal.).

At Factory B, conditions were less satisfactory—the milk supplies differed considerably in fat content and the receiving vat was small (1,000 gal.). The composition of the milk in the receiving vat could therefore change quite rapidly, and one sampling would give a much less accurate estimate of the composition of the milk required to fill one cheese vat than it did in Factory A.

A control chart for Factory B is shown in Fig. 4. As five vats of cheese were being made each day during the period concerned, one day's make was taken as a sample. The variation of control limits from the average F.W.F.S. was 0.78 for samples of 5 vats or 1.75 for individual vats. The safety margin allowed during this period (1.55%) was quite adequate as only one vat out of 295 showed less than 52% F.W.F.S. This margin, however, was 0.6% higher than that found satisfactory at Factory A. Installation of a larger receiving vat would have made it possible to bring the average F.W.F.S. rather closer to the 52% minimum.

Provided that a receiving vat with a capacity 2 or 3 times that of a cheese vat is used, the rapid method of standardization described can give very satisfactory results. Even with the most accurate standardization, however, it is necessary to allow a safety margin of at least 1% between the average F.W.F.S. of the standardized cheese and the minimum permitted.

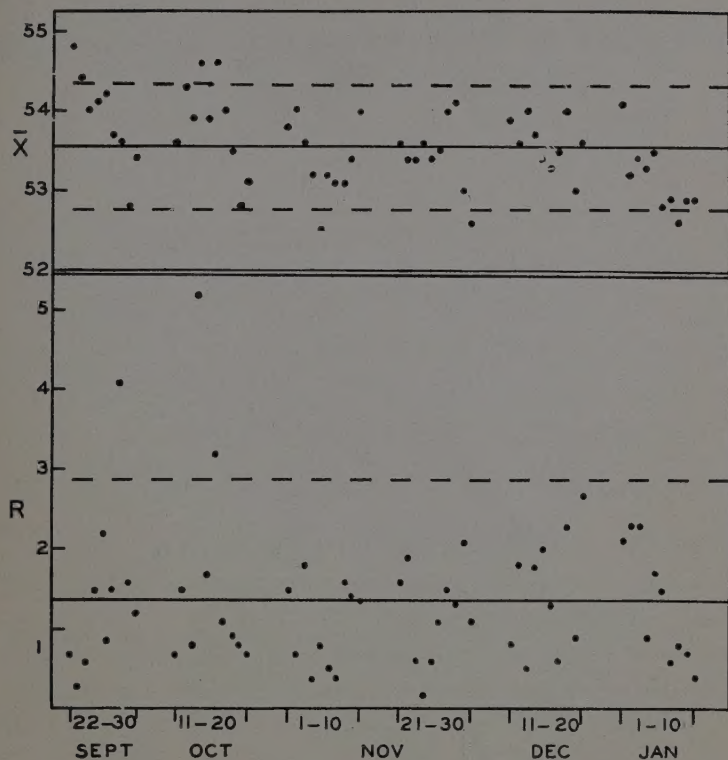


Fig. 4.—Control chart for Factory B.

$$M_x = 53.55$$

$$\bar{R} = 1.36$$

$$\text{Limits for } \bar{X} \text{ (average of 5 vats)} = 53.55 \pm 0.78$$

$$\text{Limits for individual vats} = 53.55 \pm 1.75$$

$$\text{Limits for } R = 0, 2.87$$

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